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Agricultural
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North Central Region
Metabolism and
Radiation Research
Laboratory

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Fargo, North Dakota
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February 1, 1983

SUBJECT: Mating Types In Screwworm Populations? Comments on the
Rebuttal by Richardson et al (Science 218:1142-1145)

TO: R. A. Bram

The December 10, 1982 issue of Science (Vol. 218:1142-1145) contained some comments from ARS scientists pertaining to a screwworm article published by Richardson et al in January 1982. The ARS comments were followed by a rebuttal by Richardson et al. In early December 1982, you requested that I coordinate an "in house" analysis of the Richardson et al rebuttal. Accordingly, I requested comments from all ARS co-authors in my letter of 8 December 1982. I have discussed this matter with all 7 of the ARS co-authors. Drs. Gagné, McInnis, Whitten and myself have made comments and these are attached. The other authors felt that the Richardson rebuttal was "the same old stuff" and elected not to comment.

Based on the views of Drs. Gagné, Whitten and McInnis, my personal appraisal, and the appraisal of other ARS authors, I certainly see no reason for ARS to alter its position as originally expressed in Science.

A number of ARS scientists are currently studying screwworm populations from central and southern Mexico and using genetic, cytogenetic, behavioral, morphological and ecological techniques. I firmly believe that the possibility of reproductive isolation among various screwworm populations is a critical question that is extremely relevant to strain development and utilization. However, more data is definitely needed before this question can be answered.

L. E. LaChance

L. E. LaChance
Research Geneticist -
National Technical Advisor
for Insect Genetics

cc: A. C. Bartlett
R. J. Gagné
✓ O. H. Graham
D. O. McInnis
J. A. Seawright
C. J. Whitten

Comments on Rebuttal by Richardson et al in Science 218:1143-1145 (Dec. 10, 1982).

224
Submitted by L. E. LaChance, Metabolism and Radiation Research Laboratory, ARS, USDA.

I found the comments by Richardson et al relatively mild and extremely evasive. A number of points have been admirably dealt with by Drs. Gagné, Whitten and McInnis in their memos (see enclosures). I will not cover the same territory, but do have a few comments.

1. It is interesting that Science insisted that we edit and shorten our technical comments on several occasions and further insisted that we change the title before they would agree to publish the material, yet permitted Richardson et al to use more space than allotted to us (35 column inches vs. 46 1/2 column inches) and to include 2 pictures. I suspect there was a bias in what Science wanted to publish.

2. In their rebuttal, Richardson et al go to considerable lengths to discredit the methods and results obtained by McInnis. Yet they used the same methods, the same tissues, and larvae from the same egg masses. They seem to be saying "McInnis can't do it, but we can." Naturally, this is the same story we've been hearing for some years and reminds me of the old question, "When two people are holding hands, how can you tell whose palms are sweaty?" They list a long litany of examples where they feel McInnis erred. Dr. McInnis covers a number of points in his letter. In my opinion, their comments are mostly "smokescreens" and not really worthy of debate. For example, Richardson admits that "another error contributing to the high variance occurs in the classification of X chromosomes" and "the measurement of partially condensed chromosomes inflates the variance." I agree with both statements, but fail to understand why they feel McInnis is always wrong and they are always right.

3. One new item that surfaces in their rebuttal concerns the existence of diploid cells with "more than the usual complement of DNA." This was originally reported by Gay et al in 1970 but contested by van de Flierdt in 1975 (reprint enclosed). Thus, it seems that the "polynemy" question is still open and not as widely accepted as Richardson wants us to think. There is no indication that "polynemic diploid cells" occur in screwworms. This would require UV absorption measurements of individual cells. Since no one has done this type of analysis, we simply cannot know if such cells exist. Their claim is tantamount to saying we exclude all polynemic cells from our sample but McInnis does not. However, they provide no information on how they differentiate the two types of cells nor do they prove that polynemic cells exist. When I see the numerical data on UV absorption for screwworm brain ganglia cells compared with other types of cells, then I will be convinced--not before.



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Northeastern Region

Systematic Entomology Laboratory, USDA
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December 22, 1982

SUBJECT: Comment on Mating Types in Screwworm Populations and rebuttal by
Richardson et al. Science 218: 1142-5.

TO: L. E. LaChance

From the comments about genitalic morphology, a person reading only Science could not judge for himself the merits of either articles because the points of contention are only bare-bones statements. So Richardson et al., p. 1143, paragraph 2, write that their observations of general morphology do not jibe with those of Gagné and Peterson (1982). So one says something is white, the other says it's black. Richardson et al. state that the genitalia of a 1-day old male corresponds to a mated type (actually only in the fact the epiphallus is vertical instead of horizontal) and all the individuals of that sibship look alike. I need to know how Richardson et al. determined that the flies were 1-day old and how many specimens were searched for this character. Fig 1B shows "virgin" genitalia from a 8-day old male. It's true that that is unlikely, but were they caged with females? If they were, were female spermathecae squashed to see if they contained sperm? Again, we don't know how many genitalia of this sibship were studied. Nowhere in the articles by Richardson et al. on this subject do they give such information. Gagné and Peterson (1982), copy attached, do indicate provenance of specimens, detailed methods, and numbers examined. Our results were not ambiguous as Richardson et al. contend they were. The genitalia of ^{male} exiting from puparia are similar; wear and tear is variable within a sibship.

RAYMOND J. GAGNÉ
Research Entomologist

Enclosure

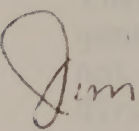
31 Jan. 1983

Dr. Leo LaChance

Senior Author of "Mating Types in Screwworm Populations?"

This letter is in response for "in-house" comments on the response of Richardson et al. to the technical comments assembled by ARS scientists for Science.

I feel that we should be aware that both the ARS authors and Richardson et al. were correct in their reporting of the sterility percentages of egg masses recovered in the 2nd phase of the Arriaga test. Richardson et al. considered the sterilities among all (8) sheep pens within the release zone, whereas the ARS authors referred to the sterilities among 3 (of the 8) pens that were purposely located near the coast to reduce the effect of migration of native females from outside the release zone. The more elongate release zone (compared to previous evaluations) in the Arriaga test was necessitated by the proximity of the mountains to the coast. This factor confounds interpretation of the resulting sterility.


C. J. Whitten
Research Entomologist



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Western Region

California-Hawaii Area
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January 13, 1982

SUBJECT: Response to Rebuttal by Richardson et al
in Science (Dec. 10, 1982)

TO: Dr. Leo E. LaChance
USDA-ARS
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State University Station
Fargo, North Dakota 48105

I would like to respond to your request for USDA in-house review of the rebuttal by Richardson et al to our joint response, which was made in turn to the original Science article by Richardson et al. I hope that these comments will set the record straight on my point of view, and help provide the USDA with information to proceed with the important next phases of screwworm genetic research. I will limit my comments to those areas that most concern me, principally the cyto-genetic data. I agree that further public airing of our views in Science would not serve any useful purpose. As for myself, I will send a copy of my comments directly to Richardson and coworkers.

Unfortunately, though not surprisingly, Richardson et al did not see fit to provide, or at least acknowledge the need for, numerical chromosome data which justifies their gamodeme types. The scientific community should not have to take what they claim on faith only. Of course, having worked very closely on the same subject with Richardson and coworkers and gathered contradictory data of my own, I have other reasons for not accepting their arguments. I attempted several times to try fitting, though not forcing, my raw data to their gamodeme model, but each time the result was the same--the gamodeme model was incompatible with my data. Richardson et al refer the reader to consult themselves and their 1980 Annual Cooperative Agreement Report but this attempt to legitimize their gamodeme model is weak and belated. Why couldn't they have published complete numerical data originally, showing levels of variation between and within alleged gamodeme types? Also, even if readers obtained the 1980 Annual Report, they would notice several major changes that Richardson et al have made since then, including the elimination of one of their original 10 gamodeme types. Basically, I feel that Richardson et al's claims cannot be taken seriously until they or others provide bona fide data that may be critically evaluated by the scientific community. Meanwhile, the USDA has not been waiting by passively, as we have proposed, and successfully defended before peer reviewers, an alternative interpretation of the chromosome data (McInnis 1981; McInnis et al, accepted for publication 1982).

The coefficients of variation reported by LaChance et al for screwworm chromosome data are accurate reflections of inherent measuring errors. These errors would have been greater had we not measured chromosomes with a sophisticated electronic planimeter. Richardson and coworkers adopted this measuring device after I introduced them to it, so I believe they should have found similar measurement errors.

Richardson et al's alleged misclassifications of chromosomes by myself are wholly unsupportable. Without my raw data before them there is no way the U.T. scientists or anyone else could erringly judge my scoring decisions and vice versa. It is extremely risky using photographic prints to make subtle judgments of the kind deciding between some chromosome types 4 and 5, or some chromosome 5 types and chromosome 2. To guard against the ever present problems of overlapping chromosome arms or 3-dimensional twists of chromosomes, I implemented the method of tracing over the chromosomes on photographic prints while viewing the actual cell through a microscope. Furthermore, even if errors were made on scoring chromosomes 2, 4, or 5 utilizing my classification system, these would not be very important because the crucial core of chromosome variation, including heterozygotes, in screwworms involves primarily the sex chromosomes. Why didn't Richardson et al attempt to explain several cases of apparent heterozygosity for the X chromosome in the same figures they chose to cite?

As to the charge regarding normalization of chromosome lengths, Richardson et al made errors both of commission and omission. They claimed that I only used absolute chromosome lengths in my 1981 paper, yet Figs. 3-5 of that paper include data on "% of haploid genome" which are equivalent to normalized or standardized lengths. I chose to discuss chromosome differences there in terms of approximate absolute or real metaphase lengths because: (1) chromosomes are commonly reported in scientific literature using absolute lengths, and my data would allow comparisons to such prior data, especially that on screwworms published by Boyes and Shewell (1975), (2) classifications of individual chromosomes or whole genomes using standardized lengths did not change what was evident based upon absolute lengths alone, and (3) it is possible to have distinct genomes be recognized based upon total absolute genome length, yet appear identical in terms of standardized lengths only. Richardson et al conveniently omitted mention of my use of standardized or normalized chromosome lengths in McInnis et al (accepted for pub., 1982), specifically done to allow a direct comparison to data of Richardson et al. Normalizing chromosome lengths does help reduce variation due to cell stage or even polynemy, but for reasons stated above, I feel that the best system is to use both absolute and standardized lengths.

As for variation in chromosome length due to differences in cell polynemy, I have several comments. First, it is only ad hoc speculation on the part of Richardson et al that polynemic chromosomes are to be found in screwworms. Where are the data from screwworms to prove it? Once again, I feel they have made an unwarranted extrapolation from some other taxa to screwworms prematurely. Second, even if chromosome polynemy occurs in screwworms, it would be irrelevant to the detection of heterozygotes since all chromosomes in a particular cell would be of equal polynemy. Therefore, the authenticity of heterozygotes I have reported are not challenged by speculation on polynemy. Third, as discussed above, since I also have used standardized chromosome lengths, polynemy is again irrelevant.

Regarding the need for proper cytological techniques, Richardson et al implied that my technique was poor since I reported variation in X chromosome fluorescence. This implication is both unfounded and hypocritical. I am well aware of limitations for the various techniques involved, and these are the same

shortcomings which Richardson and coworkers faced as well. Surely they can't be equating poor slide preparation technique to all variation in X chromosome fluorescence since they, too, have reported such distinct variations as characteristic of some of their gamodemes.

Regarding the use of prophase cell stages, I again feel that Richardson et al were hypocritical. The number of clear, dividing cells in larval brains used by both the U.T. scientists and myself was so rare that I'm sure Richardson and coworkers were not so choosy. Indeed, the apparently less than fully condensed chromosomes in their Fig. 6(A) (Science, Jan. 22, '82, p. 365) exemplifies this. The continuous nature of mitotic stages suggests that there is no clear-cut separation of 'good' or 'bad' preparations, but only that some are slightly better than others.

The claim of Richardson et al that I analyzed "irrelevant features of the karyotypes" is pure nonsense. The 3 factors used by both the U.T. scientists and myself for characterizing chromosomes were: (1) standardized chromosome lengths, (2) chromosome arm ratio, and (3) chromosome fluorescence pattern. We have not differed in the bases for data interpretation, but rather only in the interpretations themselves. I deliberately followed the basic techniques of Richardson and coworkers to rule out all but differences in interpretation were they to occur.

I have the following remarks to make on other aspects of the rebuttal which particularly concerned me. The claim of Richardson et al that they couldn't get specific hybrids from us at Mission is flatly erroneous. Dr. Jim Whitten and I sent Richardson and coworkers numerous samples, including many involving F_1 hybrids of specific crosses. I should add that Jim and I received only token appreciation from the U.T. scientists for our service. An example of this kind of 'appreciation' appears in the statement by Richardson et al that they "fortuitously" obtained the hybrid of Fig. 6(B) (Science, Jan. 22, '82, p. 365). In fact, that sample along with thousands of others sent to Richardson and coworkers came through the diligent efforts of the USDA and the US-Mexico Joint Screwworm Commission. Furthermore, because I had found an unusual heterozygote, I alerted the U.T. group to the specific sample in which they found the triploid individual of Fig. 6(B).

I turn now to comment on several aspects of various field evaluations of screwworm strains. Richardson et al claim that 64 days elapsed between the end of the Chiapas low-release rate test and the onset of sterile fly (V-81) releases in zone A (see Fig. 1 from Science, Dec. 10, '82). In fact, there was only a 3-week lag before egg mass sterility rose to 10-15% in zone A, no doubt due in part to movement of sterile flies from adjacent fly release zones. I don't believe that a 3-week, or even a 9-week, lag time would be sufficient to account for the scope of gamodeme frequency changes envisioned by Richardson et al. The unidirectional wind direction diagrammed by Richardson et al in Fig. 1 above is only half-true - they conveniently ignored the evidence from the original Chiapas test which indicated that roughly equal and considerable fly movement occurred in both directions based upon sterility levels in the middle, 'control' zone B. It was just because of this evident high vagility of screwworms that ARS moved test sites for the V-81 strain evaluation to a modified zone A and included 3 relatively insulated sheep pens along the Pacific

coast. It was at these coastal, insulated pens that sterility rates reached 60% and were climbing after 4 weeks of releases. The explanation given by LaChance et al in our Science response, though brief, remains accurate. Richardson et al took our comments out of context and provided another ad hoc explanation. I have a manuscript written on this subject of the V-81 strain evaluations which I hope to publish soon. Also, I noticed that Richardson et al didn't comment on our contentions regarding the successes of the DE-9 and Sinaloa strain tests, in spite of the great genetic mismatches postulated by Richardson et al based upon their gamodeme distribution map shown in their original Science article. Those test results are strongly incompatible with the notion of severe reproductive isolation existing in screwworms, though, of course, weaker isolating forces, including mating preferences, may be present.

Finally, I would like to say that the concluding remarks of praise for the USDA by Richardson et al regarding factory strain policy, though only token, are nonetheless welcome. At least they didn't take credit for implementing those changes. I have long maintained that the program recommendations of Richardson et al and myself have been very similar, though for different reasons. Mine have been basically made as a precaution against potential genetic problems, while theirs have been stated as being extremely necessary to attain complete program success.

Don McInnis

DON McINNIS
Research Geneticist

No Multistrandedness in Mitotic Chromosomes of *Drosophila melanogaster*

Kathrin van de Flierdt

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Abstract. Feulgen cytophotometric measurements of neuroblasts in the first and third instar larvae of *Drosophila melanogaster* reveal the same DNA content for metaphases with chromosomes of different size. The total absorbance of all measured metaphases gives the four-fold value of that of the spermatids. Accordingly there seem to be no reasons to retain the assumption of a multistranded structure for the large chromosomes of metaphases in the third instar larvae.

Introduction

The discussion of strandedness in regard to chromatids has been decided of late in favor of a single-stranded model (Swift, 1973). Unexplained in this context are the structural significance of the split in anaphase chromosomes and all those cases, in which differences in DNA content could be demonstrated in diploid metaphases of the same genome. In neuroblasts of *Drosophila melanogaster* Rudkin (1963) was able to show that metaphases from the third instar larvae contain four times the amount of DNA found in the first instar larvae. Gay *et al.* (1970) have investigated this phenomenon more closely with the aid of Feulgen cytophotometry. From these studies a relation has been shown to exist between the DNA content of sperms and of metaphases from the ganglionic cells of first instar larvae, which can be interpreted by a 1c:4c ratio. The larger metaphases from the brains of third instar larvae contained, nonetheless, twice the amount of DNA as compared to those of young larvae. These must then be attributed to the 8c class.

These findings are explained by Rudkin (1963) and Gay *et al.* (1970) on the basis of different strandedness of the chromatids. The mitotic chromosomes of third instar larvae are considered to be polynemic in contrast to chromosomes of young larvae.

This hypothesis is seemingly supported by an older cytological finding. Kaufmann (1934) observed in anaphases of neuroblasts of adult *Drosophila* larvae split chromatids which were evaluated as indicative of polynemy. For Sorsa (1973) polynemy, based on cytophoto-

metric data, is the basis of discussion for the multistrandedness of mitotic chromosomes from *Drosophila* as demonstrated by his whole-mount technique.

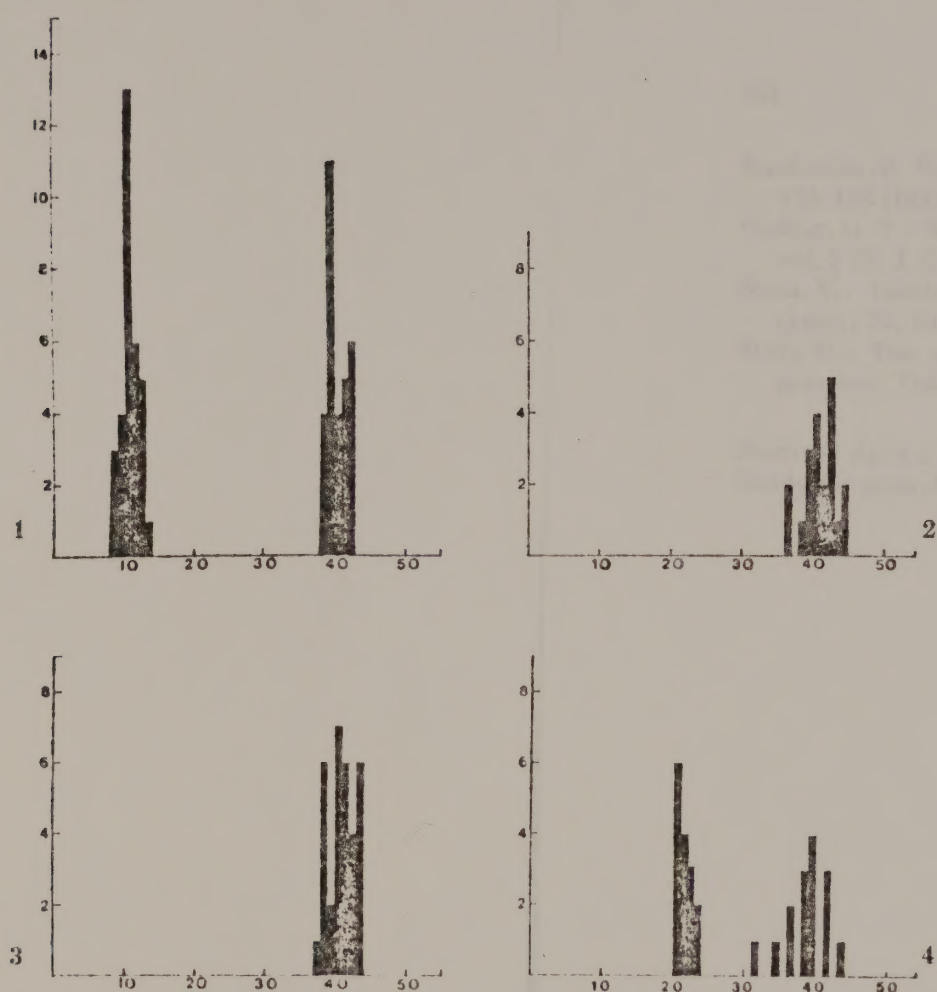
Methods

For the purposes of investigation the same Swedish-b-stock of *Drosophila melanogaster* was utilized as in the cytophotometric studies of Gay *et al.* (1970). In preparation and staining the methods of Gay *et al.* (1970) were used. Measurements of Feulgen absorption then were done at 560 nm with a Universal-Mikro-spektralphotometer from Carl Zeiss, Oberkochen.

Results and Discussion

Measurements were carried out on spermatids, interphases and metaphases of the brain cells of first instar larvae and on metaphases of brain cells of third instar larvae as well as metaphases from leg discs of the third instar. These measurements are reproduced in the diagrams of Figs. 1-4. The metaphases from the brain cells of the first and third stages show the same differences in size as those of the chromosomes depicted in Gay *et al.* (1970). Within the same brain the size of the metaphases varied greatly. The measurements of metaphases from the brain cells of third instar larvae (Fig. 1) with a value of 40.45 ± 1.03 displayed no significant difference in comparison to values of 41.55 ± 2.11 in metaphases of the first instar larvae (Fig. 2) and of 41.13 ± 1.32 for metaphases from the leg discs of third instar larvae (Fig. 3). The interphases from brain cells of the first larval stages provided values grouped around 21 and 40 (Fig. 4). As a result they are in full agreement with metaphase values. The total absorbance of spermatids (Fig. 1) is recorded as 10.73 ± 0.9 . In light of the foregoing it is proved that none of the investigated metaphases contained more than the four-fold DNA content determined for spermatids.

From these results it is obvious that the size differences of metaphase chromosomes from brain cells of *D. melanogaster* are not based on different DNA content, but rather due to different contraction conditions of the chromatids. The facts presented above allow no assumption whatsoever of a multistrandedness in chromosomes of the third instar larvae. The differences in DNA content shown up till now in the measurement of metaphase chromosomes most probably result from methodological causes. Personal investigations have revealed that because of the small size of their nuclei the DNA content of metaphases and interphases of first instar stages can only be accurately measured with a microspectral photometer when the method of complete scanning is applied.



Figs. 1—4. Integrated absorbancies in arbitrary units from different larval tissues. Fig. 1. Spermatids of larval testes, $\bar{x} = 10.73 \pm 0.9$; Ganglion cell metaphase of 3rd instar larvae, $\bar{x} = 40.45 \pm 1.03$. Fig. 2. Ganglion cell metaphases from 1st instar larvae, $\bar{x} = 41.55 \pm 2.11$. Fig. 3. Metaphases of leg discs from 3rd instar larvae, $\bar{x} = 41.13 \pm 1.32$. Fig. 4. Interphase stages of ganglion cells of 1st instar larvae. — Abscissa: Integrated absorbance. Ordinate: Frequency

Acknowledgements. Thanks are gratefully extended to Prof. Dr. H.-G. Keyl for his help in conception and execution of this study. This work was supported by the Deutsche Forschungsgemeinschaft.

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Subject: Swormlure as an attractant

Date: Feb. 3, 1983

To: MVZ. MOISES VARGAS TERAN
DR. H. C. HOFMANN
ENTOMOLOGOS ASESORES

The data presented below were obtained from a 10-day trial on Swormlure conducted in Sinaloa, Mexico. There are two maps attached to this report, one shows the marked fly release point and the trap sites, the other displays the grid where sterile flies were being dispersed at the time of the 10-day trial as well as other related information.

Our work was centered on an attempt to replicate an earlier field test conducted in the state of Guerrero in December of 1982. We were highly successful in laying the groundwork and in our day to day operations. However, some variation factors between the Sinaloa and Guerrero areas at the time of the trials should be noted:

Sinaloa - heavy precipitation on and off before and during trial period

- much greenery and heavy concentrations of moisture on the ground
- temperatures varied from the low 70^o sF to the mid 80^o sF in any given 24-hour period during our stay
- terrain was fairly level and readily accessible
- sparse population near the trap sites
- no problems with insects noted (specifically ants)
- largely an agricultural area but also a moderate abundance of livestock

Guerrero - no precipitation before or during the trial period

- less greenery and very low moisture concentrations on the ground
- temperatures a bit more extreme, varying from the low 70^o sF to the low 90^o sF during the trial period
- terrain was hilly making it less accessible for our purposes
- densely populated areas near some trap sites
- insect (ant) problem in some of the traps
- less agriculture but more livestock

On Monday, January 17, 1983, at 0900 hrs., we ground-released a total of 48 boxes of flies, 40 of which contained marked flies. The boxed flies had arrived in Mazatlan earlier that same morning via truck from Guadalajara. The swormlure used to bait the traps was part of the same swormlure used in Guerrero and we had brought it in with us three days before.

The brief description below reflects the basic set-up required for the 10-day field test.

Bait: Swormlure 4

Set-Up: 24 wind-oriented traps, 6 in each of four different directions leading from the release point(see attached map)
in each direction, traps approximated from release point as follows: 1 at 1 kilometer; 2 at 2 kilometers(400-500 meters apart); 3 at 3 kilometers(300-500 meters apart)

Data obtained from random samples(4 boxes) taken at the time the flies were released is included below.

Box #	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>
Dead at Release	3	5	5	3
Emergence	880	848	837	809
Non-Emergence	126	129	159	143
Emergence after Release	<u>28</u>	<u>30</u>	<u>37</u>	<u>34</u>
Pupae	1,006	977	996	952

Released 48 boxes(only 40 contained marked flies)
Pupation 1/5/83
Irradiation 1/11/83
Emergence 1/14/83
Release Date 1/17/83

Total Pupae	39,310	or	982.75	per box
Total Emergence	33,740	or	843.50	" " (85.83%)
# Dead Flies at Release			4.00	" " (0.47%)
# Flies Emerged after Release			32.25	" " (3.82%)

Total fly collection on a by-day basis:

Day	# of Traps In Use	Screwworms		Other Blowflies
		o	o +	
1	21	12	64(3)	50
2	23	3(3)	32(6)	28
3	6	4(3)	10(7)	4
4	23	10(2)	31(11)	48
5	23	21(1)	54(9)	13
6	24	46	192(14)	18
7	23	54	227(3)	32
8	22	29	150(2)	17
9	24	20(1)	129(1)	19
10	<u>24</u>	<u>32</u>	<u>112(4)</u>	<u>14</u>
	213	231(10)	1001(60)	243

# Flies per Trap - Male	1.08
# Flies per Trap - Female	4.70
# Marked Flies per Trap - Male	0.05
# Marked Flies per Trap - Female	0.28
# Flies per Trap - Other	1.14

NOTE: The numbers shown in parenthesis, (#), reflect marked flies and are already included in the totals.

The results below show at what distances from the release point flies were captured.

Perimeter 1 (1 kilometer from release point)

Male Screwworm Flies	73	or	31.60%	of total catch
Female Screwworm Flies	250	or	24.98%	of total catch
Marked Male Screwworm Flies	8	or	80.00%	" " "
Marked Female Screwworm Flies	39	or	65.00%	" " "

Perimeter 2 (2 kilometers from release point)

Male Screwworm Flies	66	or	28.57%	of total catch
Female Screwworm Flies	294	or	29.37%	" " "
Marked Male Screwworm Flies	0			
Marked Female Screwworm Flies	7	or	11.67%	" " "

Perimeter 3 (3 kilometers from release point)

Male Screwworm Flies	92	or	39.83%	of total catch
Female Screwworm Flies	457	or	45.65%	" " "
Marked Male Screwworm Flies	2	or	20.00%	" " "
Marked Female Screwworm Flies	14	or	23.33%	" " "

These trials were conducted by Angel Soto, biological technician from Mexico City, and myself.

Lizandro Gonzalez
Biological Technician

Distribution: W. H. SUDLOW, DR. HAROLD BROWN, DR. HUGH GRAHAM,
DR. ANGEL SOTO

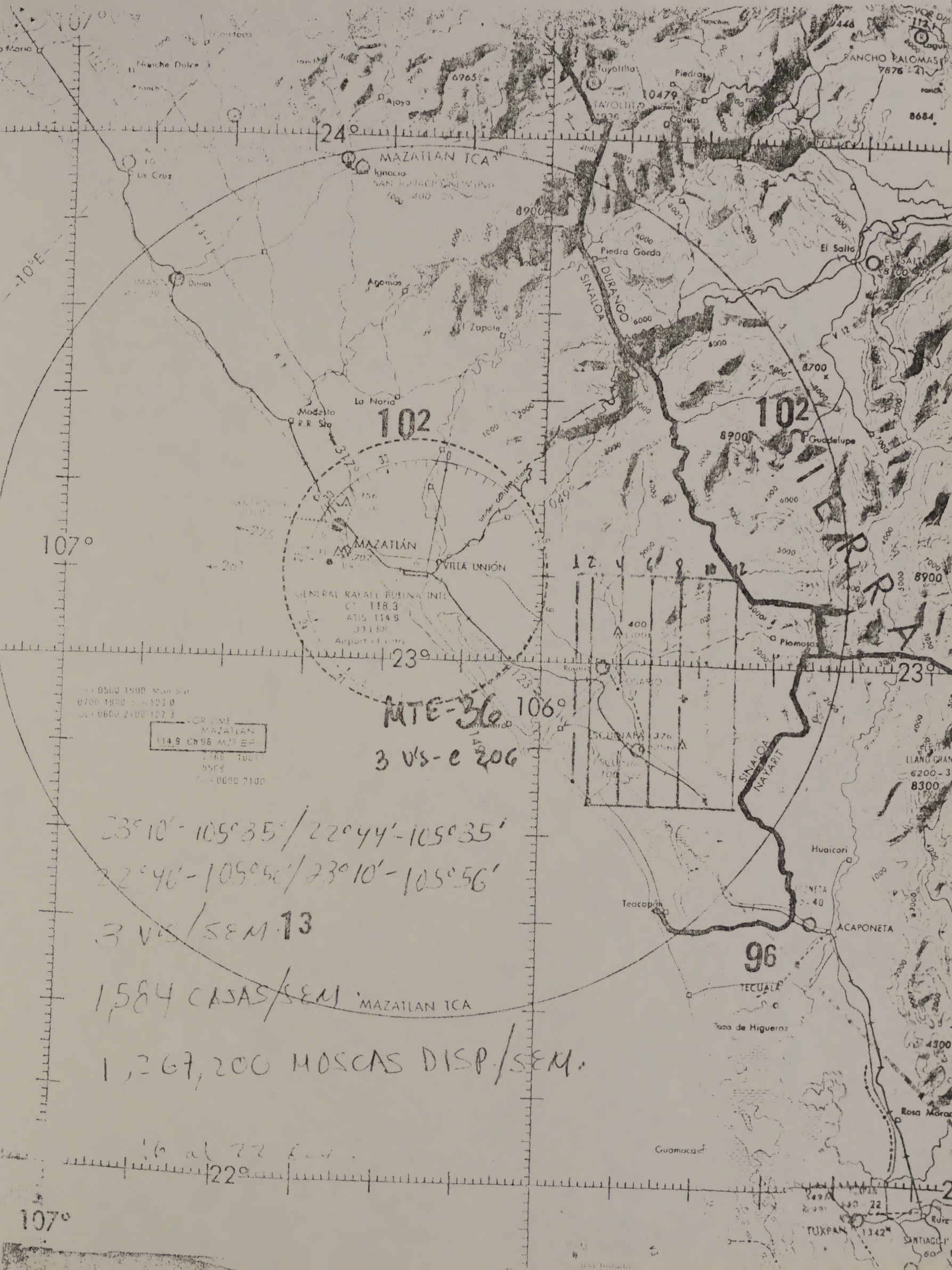
SCREWORM COLLECTION

Day #	1		2		3		4		5		6		7		8		9		10	
Trap #	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F
1	0	0	0	2(2)	4(3)	9(7)	0	0	4(1)	10(7)	4	15(7)	5	28	2	9	4(1)	24(1)	3	18(2)
2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
4	0	0	0	2(2)	0	0	0	0	0	0	0	5	1	1	0	2	0	2	2	2
5	0	4	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	1(1)
6	0	1	0	0	0	1	0	0	0	0	2	4	5	9	2	7	1	2	0	1
7	0	2	0	2(1)			0	1	0	0	2	3	1	6	0	1	0	0	0	0
8	0	2(1)	0	0			0	0	0	0	0	0	0	0	0	0	0	0	0	0
9	0	6	0	1	other		2	3	6	13	2	15	-	-	3	17	4	16	0	2
10	0	1	0	0	traps		0	0	0	1	0	3	0	1	0	0	0	0	0	0
11	0	1	0	0	not		1	1	1	4	0	0	0	0	0	0	0	0	1	0
12	1	4(1)	1(1)	1	checked		1	0	1	3	0	1	0	0	0	0	0	0	0	0
13	2	2	0	0	because		0	0	0	2	0	1	2	11	-	-	1	0	0	2
14	0	0	0	0	of		0	0	0	1	1	6	2	9	1	5	0	5	0	1
15	0	0	0	2	heavy		1	6	0	1	0	5	1	20	1	11(1)	1	5	2	6
16	1	10	0	2(1)	rain		0	1	1	2	2	12	7	24	0	4	2	12	3	13(1)
17	1	3	0	1			0	0	0	2	3	19	3	22	0	0	0	0	0	3
18	5	24	0	16			-	-	-	-	0	14(1)	6	39	1	18(1)	2	32	12	32
19	2	4(1)	1(1)	1			4(2)	9(6)	3	6(1)	13	32(2)	8	16(2)	-	-	2	11	6	23
20	0	0	0	0			0	0	0	0	4	11	8	20(1)	14	31	2	18	1	5
21	0	0	-	-			0	7(4)	1	1	8	25	0	5	0	12	0	0	0	1
22	-	-	0	1			0	0	0	0	2	2	0	2	0	0	0	0	0	1
23	-	-	0	0			0	0	0	0	0	0	0	0	1	2	0	0	0	0
24	-	-	1(1)	1			1	3(1)	4	8(1)	3	19(4)	4	14	4	31	1	2	1	1
Total	12	64	3	32	4	10	10	31	21	54	46	192	54	227	29	150	20	129	32	112
*		(3)	(3)	(6)	(3)	(7)	(2)	(11)	(1)	(9)		(14)		(3)		(2)	(1)	(1)		(4)

* - Denotes (#) number of marked flies captured and already included in the total

Trapping January -18 thru 27- 1983





MTE-36

3 V's - c 206

23°10' - 105°35' / 22°44' - 105°35'

22°46' - 105°56' / 23°10' - 105°56'

3 V's / SEM 13

1,584 CASAS / SEM

MAZATLAN ICA

1,267,200 MOSCAS DISP / SEM.

16 al 22 feb.

22°

107°

ID Document	317
Form 1	Report
Form 2	
Title	Method Development: Strain Change Report: A-82 Strai
# of Pages	4
Year	1982
Month	
Day	
Color?	☐
Spanish?	☐
Subject line	
Subject heading 1	fly strain
Subject heading 2	facilities
Subject heading 3	
Subject heading 4	
Description Note	
Author Info	Broach
Additional Authors	Charles MacVean
Recipient	
Sub-collection	Southwestern Eradication R
Treatment intent	
Temp CD folder	142
To be scanned?	☐
Scanning completed?	☐
To be re-keyed?	☐
Re-keying completed	☐



COMISION MEXICO-AMERICANA PARA LA ERRADICACION DEL GUSANO BARRENADOR

LA ENERGIA ATOMICA AL SERVICIO DEL CANADERO

KM. 2 CARRETERA A LA ANGUSTURA
CHIAPA DE CORZO, CHIS.

February 3, 1983

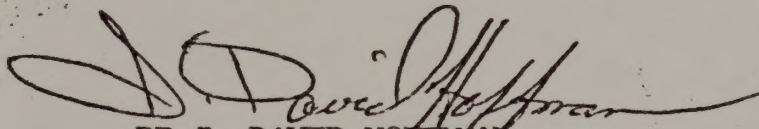
TO: DR. RONALD E. LOWE
SUB-DIRECTOR OF PRODUCTION

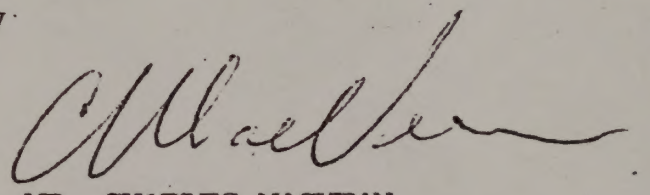
SUBJECT: REPORT ON THE REARING OF THE A-82 STRAIN

Attached is our report on the 1982 strain change. It presents the main difficulties encountered during the strain change and our recommendations for alleviating these problems. This report is thus our general proposal for modifications and additions to Methods' in-plant facilities.

We will be glad to discuss our recommendations with you and provide more specific information on equipment, facility modifications, etc.

Sincerely


DR. J. DAVID HOFFMAN
CHIEF OF METHODS DEVELOPMENT


MR. CHARLES MACVEAN
ASSISTANT OF METHODS DEVELOPMENT

c.c. Dr. Norvan L. Meyer
Dr. Robert E. Reichard
Dr. Chris H. Hofmann
Dr. James E. Novy
Dr. O. H. Graham
Sr. Raul Marroquin
Dr. Agustín Pagaza
Dr. Arturo Martinez y T.

Encl.
*svrg



United States
Department of
Agriculture

Agricultural
Research
Service

Southern Region

Cotton Production Research
P. O. Box 2131
Florence, SC 29503

February 3, 1983

SUBJECT: 1973 Report on Screwworm Nutrition Research

TO: Dr. Ralph Bram
USDA, ARS, NPS
Rm. 338, B-005
BARC-West
Beltsville, MD 20705

The subject report is enclosed.

The information on response to heat stress is in Tables 3 and 6, and figure 2. The mortality on naturally-reared flies from sheep is in Table 3, Group II. The comparison in weight and stress between strains is in Table 6. Figure 2 shows a trend to lower mortality in larvae that weight more than 68 mg. Those from sheep at an average of 95 mg would be off the graph.

I hope this will be useful. Please call me if you have any questions.

R. F. Moore

R. F. MOORE
Research Leader

Enclosure

2/s
ok

UNITED STATES DEPARTMENT OF AGRICULTURE
AGRICULTURAL RESEARCH SERVICE
Southern Region
Georgia -South Carolina Area
Southeastern Cotton Insects Investigations
P. O. Box 271, Florence, S. C. 29501

January 9, 1974

Subject: Report of Nutritional Research Conducted on Screwworm,
Oct. 29, 1973 to Dec. 12, 1973, at Mission, Texas

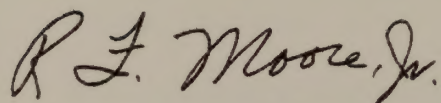
To:	W. M. Bruce, Tifton, Ga.	Lyman Henderson, Beltsville, Md.
	R. C. Bushland, Mission, Tex.	Chris Hoffman, Mission, Tex.
	E. Corrigan, Mission, Tex.	C. H. Hoffman, Beltsville, Md.
	M. M. Crystal, Mission, Tex.	W. Klassen, Beltsville, Md.
	H C Cox, New Orleans, La.	E. F. Knipling, Beltsville, Md.
	R. E. Gingrich, Kerrville, Tex.	M. E. Meadows, Mission, Tex.
	Jack Graham, Mission, Tex.	E. A. Taylor, Weslaco, Tex.

Through: H. M. Taft, Research Leader

The subject report is the result of research conducted during a temporary assignment to the Screwworm Research Laboratory, Mission, Texas.

The original goal of producing a wild type of fly with a blue color and robust appearance was not accomplished, but the more general goal of determining if some nutritional changes in the rearing might produce a better quality fly was realized. The results of the studies indicated (1) that the waste products from the larvae probably limits their size, (2) that the lower limits on the size of acceptable larvae should be 65-68 mg instead of the present 60 mg, and (3) that larval size and the mortality resulting from exposure of adult flies to heat stress could give laboratory data that would have a direct relationship to survival in the field and consequently the performance of the released flies in the eradication program.

Some additional experiments will be conducted in January and February 1974 to complete portions of the studies.



R. F. Moore, Jr.
Research Entomologist

REPORT OF NUTRITIONAL STUDIES ON THE SCREWORM
CONDUCTED AT THE SCREWORM RESEARCH LABORATORY

October 29 - December 12, 1973

R. F. Moore, Jr., Research Entomologist
Southeastern Cotton Insects Investigations
Florence, S. C. 29501

Dr. R. E. Gingrich and I were requested to review the nutrition of the screwworm in a memorandum from Dr. Waldemar Klassen dated Feb. 26, 1973. Dr. Gingrich and I visited the Screwworm Research Laboratory, Mission, Texas in early April 1973 and plans were made to conduct a series of experiments, to begin in the fall and lasting approximately one month. The experiments were to be designed to produce, from a laboratory diet, a fly with the "blue" color and "broad" thorax of the wild or animal-reared fly.

To assist in the experimental design, information on the composition of beef blood, amino acid content of blood and muscle, and an amino acid analysis of screwworm larvae and adults was requested. Dr. Klassen assisted in routing this request to the proper individuals so that the data were available for the experiments.

I arrived in McAllen, Texas on Oct. 28, 1973 and reported to the Screwworm Research Laboratory on Monday, Oct. 29. Dr. Gingrich was also there and after consultation with Dr. R. C. Bushland and Jack Graham, a series of experiments was planned.

The available data indicated that a "wild" type of fly had not been produced, even on the best diets of horsemeat, blood and plasma. Thus, it seemed advisable to attempt to make changes in the "best" diet to determine if it could be modified to produce a wild type fly. Examination of the experiments with horsemeat: blood: plasma diets revealed that the blood or plasma was always diluted with water by a factor of 1/3 to 1/2.

The reasons for the limitation on color, robustness, and larval size of insects from laboratory diets are not known. They may be related to the differences between "living" and "dead" tissue; such differences would be subtle, perhaps complex and difficult to determine. It was felt that a deficiency in some major component of the media, such as protein, minerals or vitamins, should be eliminated as a first step. Therefore, protein as egg albumin, 3 mineral mixtures, or a vitamin mixture were added to the 1000 ml of water and in addition, plasma was substituted for the 1000 ml of water to determine if dilution of one or all of these components determined the type of fly produced from the diets. Availability of food, the accumulation of wastes, and pH were also recognized as possible limitations on the type of fly and some experiments were designed to test these factors.

The original intention was to observe the incidence of blue flies and make some measurements of the thorax as an indication of the success of the modifications of the diet. It was soon apparent that there was considerable variation in the color and appearance in both lab and wild flies and that valid objective measurements of the differences would be difficult. Discussions with individuals associated with the Screwworm Project raised several questions about the color of wild flies and the factor(s) that make the wild fly appear more "broad shouldered" or robust. The incidence of "green" screwworm flies in nature, the size variation and the effect of premature pupation (forced by death of the host) on color and robustness of the flies are other unknowns.

Some individuals felt that there was a color change in the flies after death. This was tested by freezing some laboratory-reared flies, then holding some at room temperature, heating some at 37°C for several hours, and putting others in the sun. Those heated at 37° or exposed to the sun changed to the blue color of the wild fly. Flies which are caught in traps might be exposed to such conditions either in the trap or in the vehicle in which they are transported. Thus, an unknown number of green wild flies may change to blue in the trapping and handling process. Captured laboratory flies might also turn blue and be incorrectly identified as wild.

Dr. Bill Hightower had, in the past, made measurements of the width and length of the thorax of wild and laboratory-reared flies. The ratio of length/width from these data was found to be 1.02 ± 0.02 for 25 wild flies and 1.05 ± 0.02 for 25 lab flies. This is not statistically significant by the t test. Efforts to find specific characteristics to measure the "robust" characteristic were unsuccessful.

The robust characteristic was thought to be present, even in small wild flies which result from premature pupation. This was discussed with Mr. Calvin Jones and at his suggestion a field trip was made to the King Ranch at Encino, Texas. Larvae in the ears of cattle were obtained; a group of large larvae were forced to pupate without completing their feeding and produced pupae weighing 49 mg. This corresponds to a larval weight of wild flies of approximately 70 mg but would be slightly larger than the usual laboratory-reared larvae. A second group of larvae was about 24 hours old; these were forced into the horse meat diet where development was completed and pupae weighing 53.9 mg were produced. Mr. Jack Graham examined the adult flies from these larvae and compared them with flies that had been reared on sheep, on hydroponic diets, and horse meat diets. Only those from sheep had the appearance of wild flies. Thus, flies which had developed from larvae taken from cattle and forced to pupate prematurely or which completed a substantial portion of their larval development on laboratory diets could not be distinguished from flies reared wholly on laboratory diets. In other words, both blue and green flies were present and they did not have the robust appearance of wild flies. It was concluded that the blue color and broad thorax or robust appearance were difficult to define and were inadequate for evaluation of differences in flies from the experimental diets.

Since the original characteristics were lacking in objectivity, the following methods were selected for evaluation of the flies from the experimental diets: (1) Average larval weight as determined by the weight of 10 larvae which were selected just prior to pupation; (2) Tendency to fly-- this was a modification of a published method for determining the tendency of an insect to fly. Modification of the method, equipment, and initial testing had been done by Dr. M. M. Crystal. This method was applied to the males from each diet in the expectation that flies from the best diets might have a greater tendency to fly; (3) Stress survival-- Jack Graham had done some experiments with the survival of adult flies at 39°C. It was reasoned that survival under this type of stress would be a desirable characteristic and such flies should be found in the best

diets. In the test, 20 male flies that were 5 days old were exposed to 36°C, without food or water for 16 hours and the percent mortality recorded; (4) Categorizing by physical characteristics-- 3 technicians, experienced in the classification of wild and laboratory-reared flies, classified 20 male flies from each diet. A successful diet should show a larger percentage of flies appearing like the wild type.

Examination of the results of the evaluation tests of flies from the Group III diets suggested the possibility that the size of the flies might be a factor in survival under the stress of exposure, without food or water, to a temperature of 36° for 16 hours. Therefore, some measurements of the wet weights of survivors were made immediately at the end of the test. Measurements of the dry weights of the survivors and dead flies were made after drying to a constant weight at 36°C.

Five groups of experimental diets were set up. Each group contained a Standard diet which contained 4 ml formaldehyde, 1000 ml water, 500 ml blood plasma, and macerated horse meat (approximately 1500 gm) for a proper consistency. Unless otherwise indicated, the changes in the diets were to modify the 1000 ml of water by adding egg albumin, minerals, vitamins or by substitution of plasma for the water.

The diets were made up on Friday afternoon and one replicate of each diet was started on Sat., Sun., and Mon. The larvae pupated in 4 days and emerged as adults in 15 days. The adults were tested when 5 days old.

The diets were:

Group I*

1. Standard
2. 0.1% Wesson salts
3. 0.1% Human blood salt mix
4. 0.1% House salt mix
5. 20 ml of Vanderzant vitamin mix

* Dr. Gingrich determined the osmotic pressure of the salts after the tests were begun and found that 0.9% was more nearly isotonic. It was also found that the vitamins were only 1/3 of that which might be required. Thus, flies from this group were used in an effort to standardize the evaluation procedures.

Group II

1. Standard
2. Undiluted plasma
3. 6% egg albumin + amino acids*
4. 6% egg albumin + amino acids, 0.1% Wesson salts, 20 ml vitamins
5. 6% egg albumin
6. 6% egg albumin + amino acids, 5% dried blood, 0.1% Wesson salts, 20 ml vitamin mix

* Selected amino acids were added to the egg albumin to make its amino acid composition more like that of bovine plasma.

Group III

1. Standard - pH not adjusted
2. Standard - pH 7.0
3. 1000 ml citrated whole blood, pH not adjusted
4. 3% egg albumin, pH 7.0
5. 3% egg albumin + Lysine and Phenylalanine*, pH 7.0
6. 3% egg albumin + Lysine and Phenylalanine*, pH not adjusted

* The "protein score" concept was used as the basis for modifying the amino acid composition of the egg albumin. The carcass analysis of larvae reared on sheep was used as the standard and lysine and phenylalanine were the most limiting amino acids.

Group IV - all diets were pH 7.0

1. Standard
2. 0.9% Wesson salt mix
3. 0.9% Human blood salt mix
4. 0.9% House salt mix
5. 60 ml Vanderzant vitamin mix
6. 3% egg albumin, 60 ml vitamin mix, 0.9% Human salt mix

Group V - all diets were pH 7.0

1. Standard
2. 6% egg albumin
3. 6% whole egg
4. 12% cottonseed flour
5. Texturized soybean protein, no horse meat, 6% egg albumin, 0.9% Human salt mix, 60 ml vitamin mix, 500 ml plasma
6. Texturized soybean protein, no horse meat, 1000 ml of a 1:1 dilution of hydroliquid, 500 ml plasma

Results

Group II Diets.-- Substitution of plasma for the water or the addition of egg albumin in the water of dilution produced slightly larger larvae (Table 1, diet 2) than did the Standard diet (Table 1, diet 1). However, they were not any larger than some larvae from the Standard diet in other groups (Table 1, Group III, diets 1 & 2; Group V, diet 1). Flies from diet 3 were the least responsive in the flight test (Table 2) and had one of the higher mortalities in response to stress (Table 3). Flies from diet 6 were most responsive in the flight test (Table 2) and had one of the lower stress mortalities (Table 3).

Group III Diets. --Adjustment of the pH to 7.0 did not make any consistent difference in larval weights, flight response, or stress response (Tables 1, 2, 3). Addition of lysine and phenylalanine to egg albumin (diets 5, 6) to correct a theoretical deficiency of these amino acids (based on amino acid analysis of the carcass of larvae which had developed on sheep) did not produce any change in larval weight (Table 1) or flight response (Table 2) but flies from this diet at pH 7.0 did have a higher stress mortality (Table 3).

Group IV Diets. --This group of diets was expected to be the most significant of those tested because the media was adjusted to pH 7.0, the concentrations of vitamins and minerals were adjusted to a more nearly correct amount than those in Group I and some modifications in the techniques to decrease waste and increase the availability of food had been made. Examination of the larval weights, flight tendency and stress response does not show any obvious differences between the Standard and the test diets.

Group V Diets. --These diets were to test the quality of different proteins. The larger larval weights in this group are attributed to the reduced number of larvae per container and the addition of plasma to make the diet more liquid during the last 24 hours of larval development. Larvae from diets 1, 2, and 3 are comparable. Diet 4 with 12% cottonseed flour (CSF) did not support growth beyond 24 hours, reduction to 6% CSF at the end of 24 hours improved growth somewhat and a further reduction to 3% CSF at the end of 48 hours permitted larvae in replicate 3 to mature. The CSF may have some value in diets at 3%. In diets 5 & 6, the texturized soybean protein was substituted for the meat and constituted approximately 50% of the diet; no development occurred on these diets.

The data were examined by plotting flight response against larval weight (Fig. 1). The data are from 18 different diets which might explain the scatter of the points. However, the distribution of the points suggest that the larger flies do have a greater tendency to fly. The use of a single diet and refinement of the flight response test should reduce the variation and confirm this observation.

The same variations from the 18 different diets are present in a plot of the stress mortality vs larval weight. This plot (Fig. 2) shows rather clearly that the mortality is less in flies from the larger larvae.

The survival value of the larger flies is further shown in a comparison of the dry weights of the survivors and dead from the stress test. At the end of the stress treatment period, the survivors were killed by crushing and then both survivors and dead were dried to a constant weight at 36° C for 24 hours and then weighed. It was found in 23 out of 29 comparisons that the survivors weigh more than those killed by the treatment (Table 4).

The wet weight of the survivors in Groups IV and V were determined (Table 5). It is seen that the survivors average about 25 mg or more regardless of the diet or replicate.

The conditions in a wound were simulated by the apparatus in Fig. 3. It provides for a continuous flow of plasma over the larvae to remove the waste products and supply fresh nutrients. Two types of experiments were done with this apparatus. In one, the plasma was made to flow over horsemeat; in another, the plasma flowed through cotton linters. Two groups of larvae from the horsemeat and plasma averaged 84.8 mg in the first group and 90 mg in the second group with the largest larva weighing 95 mg. Those from cotton linters only plus plasma weighed 50 mg.

Summary

1. Adult flies that have been killed by freezing or other methods will turn blue when exposed to temperatures near 36°C.
2. Wild flies reared on an animal and forced to pupate prematurely cannot be distinguished from media-reared flies.
3. Fresh meat diets and fresh Hydro diets have a pH of 6.0. Diet 24 hours old and "spent" Hydro have a pH of about 7.0. Based on larval weights, adjustment to pH 7.0 of the meat diets did not seem to make any difference in their growth (Group III diets).
4. The addition of protein to the diet was the only change which seemed to affect larval size. The effect of the variables of vitamins, minerals, pH, and even the type of protein appear to be obscured by the limitations imposed by the accumulation of waste products and/or the availability of food (especially during the last 24 hours).
5. Flies from large larvae appear to have the highest response in the flight tendency test (Table 2, Fig. 1) and this may indicate that the larger flies have a greater "biological vigor".
6. The flies from the larger larvae have the lowest mortality in the stress test. Weights of survivors and dead support the idea that the larger flies are the ones that survive the exposure to heat.
7. The findings in 5 and 6 above and Figs. 1 and 2 suggest that flies from larvae that weigh 68 mg or more have a higher flight response, less stress mortality, and probably live longer in nature than those weighing less.

Miscellaneous

1. One comparison of wild flies reared on cattle with those reared in the laboratory seemed to show a different type of activity and movement. Those from the lab seemed hyperactive with movements that were jerky and excessive. There seemed to be less waste movement in the wild flies. Other observations have not confirmed this but additional observations are suggested; if the type of movement in the lab flies is confirmed, it may indicate a deficiency of thiamine and/or niacin.
2. The question of the toxicity of formaldehyde has been raised on several occasions. If it is toxic, it may be due to its formic acid content. This might be checked and if formic acid is toxic, then the formaldehyde should be stored over calcium chips.
3. It was noted that benzene is used as a killing agent in the traps. Benzene is very toxic to humans and should be used with extreme caution. It is suggested that ethyl acetate be tried as substitute.

Conclusions

The original goal of producing a wild type of fly with a blue color and robust appearance was not accomplished, but the more general goal of determining if some nutritional changes in the rearing might produce a better quality fly was realized. The results of the studies indicated (1) that the waste products from the larvae probably limits their size, (2) that the lower limit on the size of acceptable larvae should be 65-68 mg instead of the present 60 mg, and (3) that larval size and the mortality resulting from exposure of adult flies to heat stress could give laboratory data that would have a direct relationship to survival in the field and consequently the performance of the released flies in the eradication program.

Recommendations for Additional Studies

I. Stress Mortality.--The lower survival, under heat stress, of larvae that weigh less than 65 mg (Fig. 2) suggests that low survival of the released flies may have been a factor in the 1972 outbreak of screwworms in Texas.

The stress test appears to make possible the evaluation of the expected performance of the released "mass-reared" flies in the eradication program. It is suggested that first priority be given to refining and standardizing this test and relating the survival under laboratory conditions to survival in nature.

The following points of the test need to be standardized:

1. Optimum age of the fly for testing.
2. Time interval of emergence of adults.
3. Pre-treatment.
 - a. Amount of CO₂ anesthesia and recover time.
 - b. Interval between receiving food and water and the treatment.

4. Exposure temperature.
5. Length of exposure.

Relationship of survival in the lab test to that in nature might be accomplished in 2 ways: (1) Determine the survival of wild or animal-reared flies and use this data as an optimum or goal; (2) produce larvae that vary in size and determine the response of the resulting flies to laboratory stress and compare with the survival of similar flies in a standardized field cage test.

II. Nutritional Experiments. --The normal sized larvae which developed in the apparatus with horsemeat and continuously flowing plasma suggests that larval waste products limit the size of the larvae and also limits the effects of nutritional changes in the diet.

It is suggested that an effort be made to perfect an apparatus similar to that in Fig. 3 for use as a research tool. Dependable regulation of temperature and liquid flow (perhaps intermittent) through the larval medium would allow a more realistic appraisal of nutritional changes in the diet. For example, it was possible to produce 90 mg larvae with horsemeat and flowing plasma. What is the maximum size larvae that would be produced with a flow of Hydroponic fluid through the cotton linters? Such a device should be able to evaluate the quantity and quality of the ingredients in the Hydroponic diet and determine the most economical diet that would produce the best larvae because the waste products would be removed as a limiting factor. Use of such a flow-through technique would probably also reduce rearing costs by eliminating the need for removal of spent diet.

Efforts should also be made to determine what is in the waste that limits larval growth. Such information could make it possible to neutralize the wastes or remove the toxic component and reclaim the unused nutrients in the spent Hydro liquid.

Acknowledgements. --Many individuals contributed much to this research, both in actual assistance in the work and in discussions of the varied aspects of the screwworm program. Dr. Bushland, Dr. Gingrich, Jack Graham, and Frank Dutley were especially helpful in acquainting me with the screwworm program and providing basic information on the various aspects of rearing screwworms. Dr. Bushland, in addition, provided the necessary materials and facilities for the research. Thelma Jester was also helpful in securing materials and assistance. I greatly appreciate the willingness of Dr. M. M. Crystal to permit the free use of his laboratory space and equipment.

The services of Bob Handke are much appreciated. Bob was dependable in carrying out instructions, performed his duties with a minimum of instruction, anticipated the needs of the program, showed ingenuity in adapting laboratory apparatus to different needs, and he willingly worked additional hours outside the normal work schedule. I believe he is an excellent technician and should be encouraged to further increase his technical and formal training.

Table 2. Flight tendency of 5-day-old male screwworm flies; 3 replicates of 20 males each unless indicated otherwise. (Percent responding)

	Diet	Replicate			Average
		1	2	3	
<u>Group II</u>					
Standard	1	45		33	39
1500 plasma	2	45		32	39
6% EA + AA	3	40		24	32
6% EA + AA, WS, VM	4	38		32	36
6% EA	5	37		45	41
6% EA, WS, 5% DB, VM	6	53		48	51
Plant		20	33		27
Sheep		35			35
<u>Group III</u>					
Standard, pH Na	1	47	23	30	33
Standard, pH 7.0	2	55	23	17	32
Whole blood, pH Na	3	23	-	-	-
3% EA, pH 7.0	4	51	44	23	39
3% EA+Ly, Ph, pH 7.0	5	42	42	45	43
3% EA+Ly, Ph, pH Na	6	43	48	33	41
Plant		43	27	-	35
<u>Group IV</u>					
Standard	1	20	48.4	38.4	35.6
0.9% WS	2	15	41.8	47.1	34.6
0.9% HB	3	13.3	41.8	46.7	34.1
0.9% Ho	4	30	41.8	-	35.9
VM	5	18.3	36.7	46.7	33.9
3% EA, VM, 0.9% HB	6	25	38.4	-	31.7
<u>Group V</u>					
Standard	1	22	48	36	35
6% EA	2	23	27	34	24
6% Whole egg	3	20	11	34	22
12% cottonseed flour	4				
Tex. soybean protein,					
6% EA	5				
Tex. soybean protein,					
50% Hydro.	6				

Abbreviations: EA=egg albumin; DB=dried blood; VM=Vanderzant vitamin mix; AA=amino acids; HB=Human blood salt; WS=Wesson Salt; Ho= House salt; Ly=lysine; Ph=phenylalanine.

Table 3. Mortality of 5-day-old male screwworm flies after 16 hours exposure (without food or water) to 36°C. 20 males per replicate unless indicated otherwise. (Percent mortality)

Diet	Replicate			Average
	1	2	3	
<u>Group II</u>				
Standard	1	90	35	62.5
1500 plasma	2	85	100	92.5
6% EA + AA	3	75	100	87.5
6% EA+AA, WS, VM	4	60	90	85
6% EA	5	30	89	59.5
6% EA, WS, 5% DB, VM	6	55	70	62.5
Plant		100		100
Sheep		40	55	40
<u>Group III</u>				
Standard, pH Na	1	0	60	80
Standard, pH 7.0	2	15	75	72.5
Whole blood, pH Na	3	75	-	75
3% EA, pH 7.0	4	75	85	66.7
3% EA+Ly, Ph, pH 7.0	5	90	95	93.3
3% EA+Ly, Ph, pH Na		65	75	68.3
Plant		100	90	95
<u>Group IV</u>				
Standard	1	90	100	83.3
0.9% WS	2	95	95	95
0.9% HB	3	60	85	80
0.9% Ho	4	100	100	100
VM	5	95	100	97
3% EA, VM, 0.9% HB	6	80	85	82.5
Plant-TxMx		65	40	52.5
<u>Group V</u>				
Standard	1	50	85	65
6% EA	2	45	75	50
6% Whole egg	3	20	65	48
12% cottonseed flour	4		65	65
Tex. soybean protein, 6% EA	5			
Tex. soybean protein, 50% Hydro.	6			
Plant-TxMx		40	65	56.6

Abbreviations: EA=egg albumin; DB=dried blood; VM=Vanderzant vitamin mix; AA=amino acids; HB=Human blood salt; WS=Wesson salt; Ho=House salt; Ly=lysine; Ph=phenylalanine.

Table 4. Dry weights of flies that survived and died from heat stress.
(Weights in mg)

Diet	Replicate							
	1		2		3		Average	
	surv.	dead	surv.	dead	surv.	dead	surv.	dead
<u>Group III</u>								
1 Standard, pH Na					9.5	9.1		
2 Standard, pH 7.0					10.7	8.8		
3 Whole blood, pH Na								
4 3% EA, pH 7.0					10.2	10.0		
5 3% EA+Ly, Ph, pH 7.0					10.3	8.9		
6 3% EA+Ly, Ph, pH Na					9.0	9.3		
Average					9.9	9.2		
<u>Group IV</u>								
1 Standard	9.9	9.1		9.9	9.9	9.4	9.9	9.3
2 0.9% WS	9.0	8.2	8.7	8.3	9.0	8.7	8.9	8.4
3 0.9% HB	8.5	8.0	8.6	8.8	8.0	8.5	8.4	8.4
4 0.9% Ho	-	7.8		9.3		9.0		
5 VM	9.7	8.7		9.0		10.0	9.7	8.7
6 3% EA, VM, 0.9% HB	8.7	8.9		9.1			8.7	8.9
Plant(TxMx)	8.7	8.5	9.3	8.4				
Average	9.1	8.4*	8.7	8.5*	9.0	8.9*	9.1	8.7
<u>Group V</u>								
1 Standard	11.2	10.1	13.1	11.1	11.2	10.9	11.8	10.7
2 6% EA	11.4	10.8		10.4	11.1	11.3	11.3	11.1
3 6% whole egg	12.2	11.4		9.6	11.5	10.8	11.9	11.1
4 12% cottonseed flour					9.7	9.0		
5 Tex. soybean pro- tein, 6% EA								
6 Tex. soybean pro- tein, 50% Hydro.								
TxMx	9.0	8.2	9.5	8.1	9.6	8.0	9.4	8.1
Average	11.0	10.1	13.1	9.8	10.6	10.0	11.1	10.3

Abbreviations: EA=egg albumin; DB= dried blood; VM= Vanderzant vitamin mix;
AA= amino acids; HB=Human blood salt; WS=Wesson salt; Ho=House salt;
Ly=lysine; Ph=phenylalanine; TxMx=Tex-Mex strain of flies.

* Averages do not include values where 100% mortality occurred.

Table 5. The weight of male flies that survived heat stress. (Weights in mg)

Diet	Replicate			Average
	1	2	3	
<u>Group IV</u>				
1 Standard	29.1	-	22.1	25.6
2 0.9% WS	26.9	25.8	27.6	26.8
3 0.9% HB	23.1	24.8	22.7	23.5
4 0.9% Ho	-	-	-	-
5 VM	25.1			25.1
6 3% EA, VM, 0.9% HB	25.6	25.2		25.4
Plant-TxMx	23.2	25.1		24.1
Average	25.5	25.2	24.1	25.1
<u>Group V</u>				
1 Standard	33.5	37.6		35.6
2 6% EA	33.9	29.9		31.9
3 6% whole egg	35.2	30.6		32.9
4 12% cottonseed flour				
5 Tex. soybean protein, 6% EA				
6 Tex. soybean protein, 50% Hydro.				
Plant TxMx	25.0	26.8		
Average	31.9	31.2		33.5

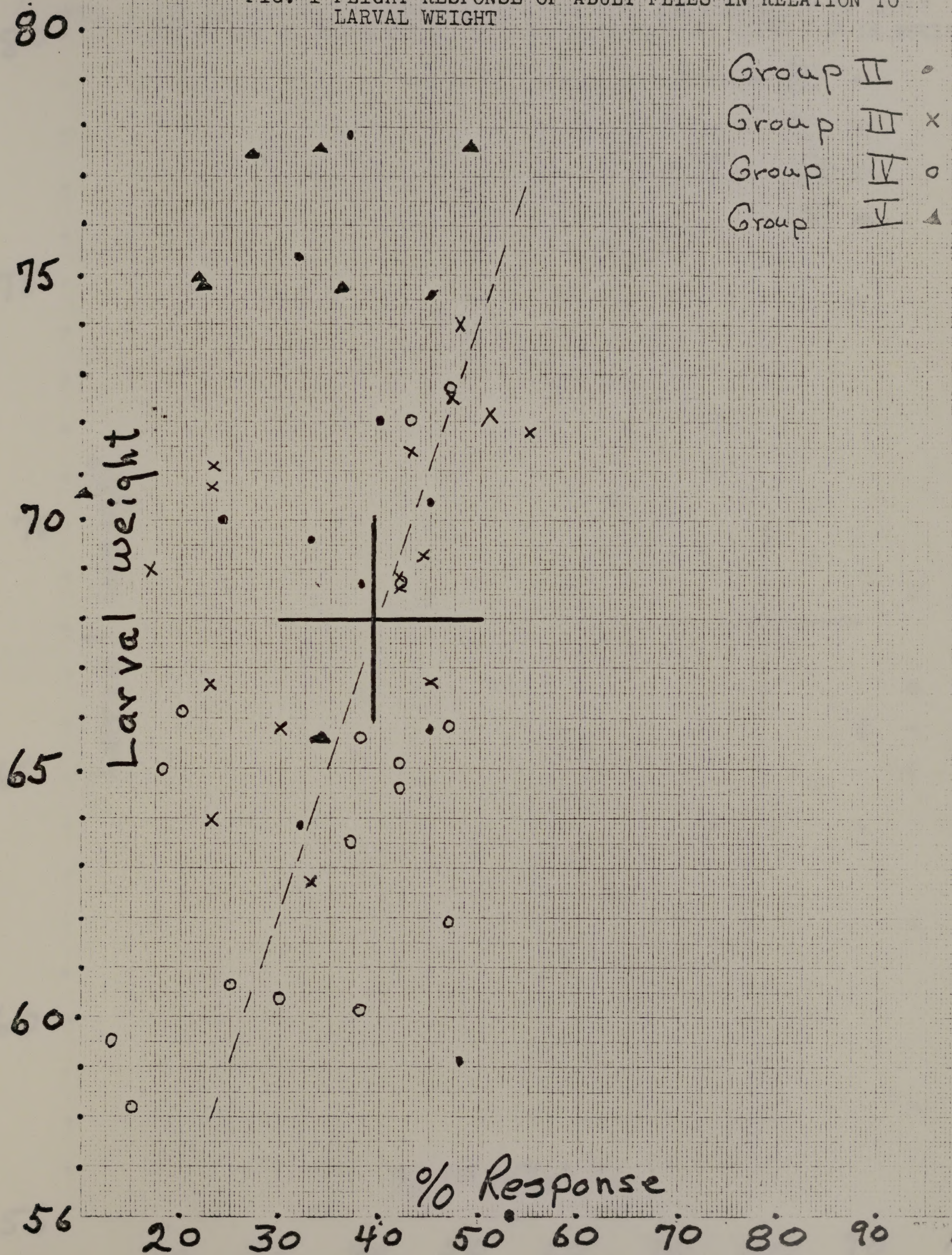
Abbreviations: EA=egg albumin; DB=dried blood; VM=Vanderzant vitamin mix; AA=amino acids; HB=Human blood salt; WS=Wesson salt; TxMx=Tex-Mex strain of flies; Ho=House salt.

Table 6. Comparison of larval weight, stress mortality, and flight response of flies from four different sources.

Source	No. of replicate	Avg. larval weight (mg)	Stress % mortality	Flight % response
Sheep - Florida strain	2	95	53	35
Horsemeat standard-Florida strain	12	70.2	70	62
Hydroponic - APHIS	3	61 *	97	31
Hydroponic - TxMx	5	61 *	55	-

* Average weight of flies being produced by the Rearing Plant at the time the tests were conducted.

FIG. 1 FLIGHT RESPONSE OF ADULT FLIES IN RELATION TO LARVAL WEIGHT



80

FIG. 2 THE RELATIONSHIP BETWEEN LARVAL WEIGHTS AND MORTALITY OF ADULT FLIES DUE TO EXPOSURE TO 36°, WITHOUT FOOD OR WATER FOR 16 HOURS

75

70

65

60

56

Larval weight

% Mortality

- Group II •
- Group III x
- Group IV ○
- Group V ▲

20 30 40 50 60 70 80 90 100

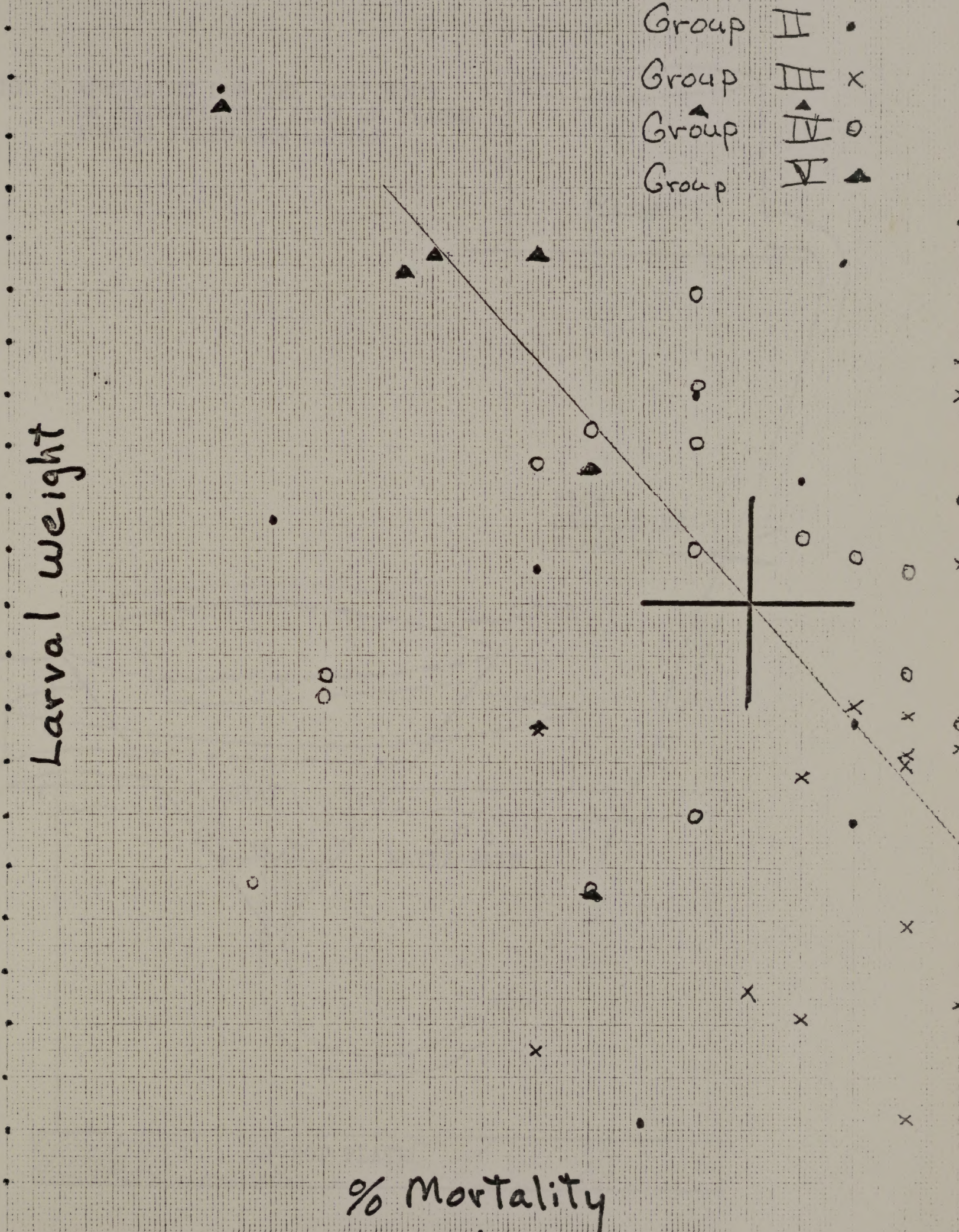
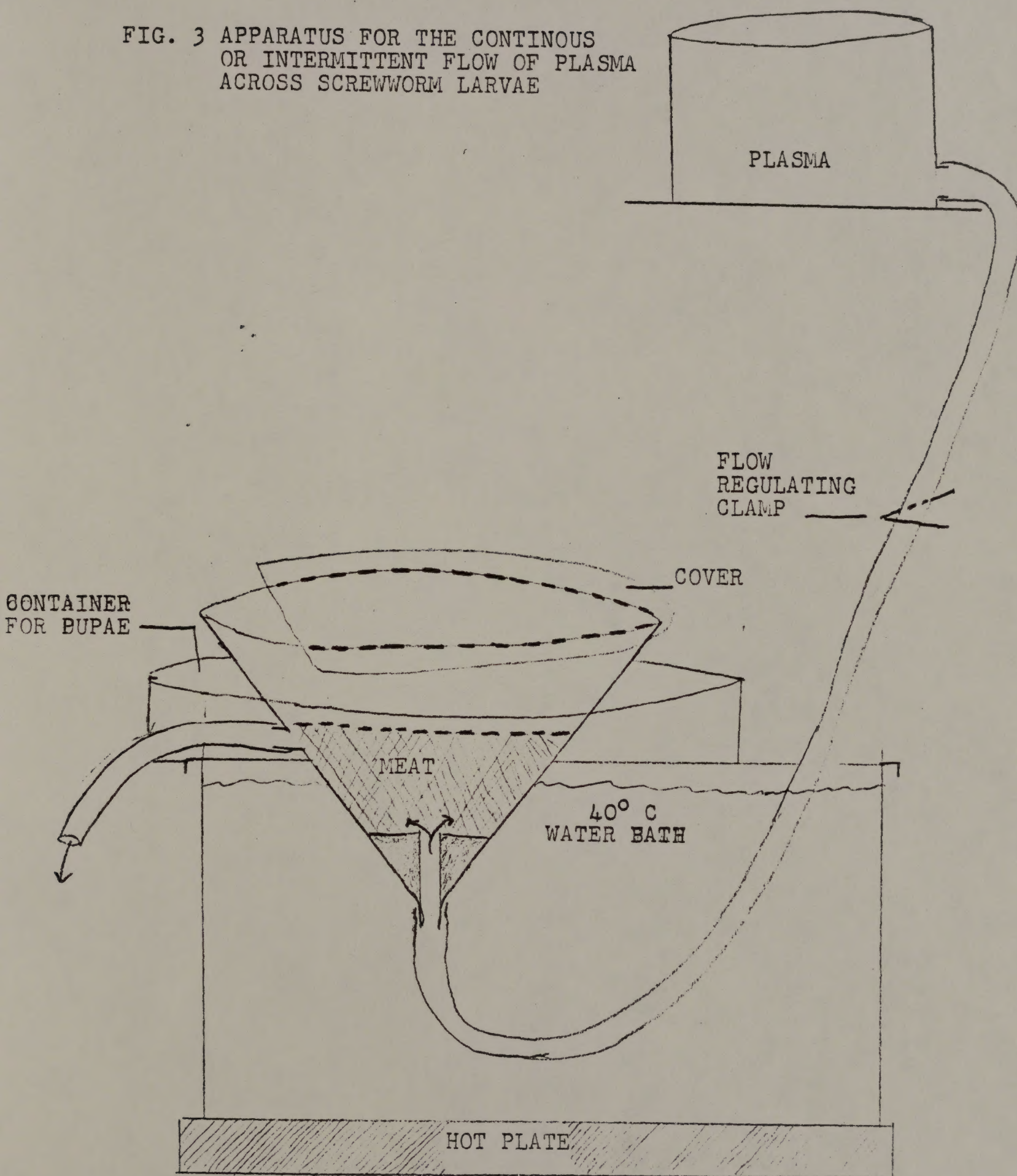


FIG. 3 APPARATUS FOR THE CONTINUOUS
OR INTERMITTENT FLOW OF PLASMA
ACROSS SCREWORM LARVAE





United States
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Administrative
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Division

6505 Belcrest Road
Hyattsville, Maryland
20782

February 4, 1983

Dr. R. J. Brenner
Research Entomologist
Screwworm Research
Apartado Postal #544
Tuxtla Gutierrez, Chiapas
Mexico

Dear Dr. Brenner,

Following receipt of your letter requesting information on ethidium bromide and propidium iodide, I checked several sources we routinely investigate for chemical toxicity data. The computerized data banks (Toxline, Toxicology Data Bank, and the NIOH Registry of Toxic Effects of Chemical Substances-RTECS) did not have much information. In fact, the attached RTECS printout was about all we could come up with from the automated sources.

I then called the Sigma Chemical Company Technical Services Office in St. Louis, (314) 771-5765, and spoke to Dr. Lee Watrud. He provided the following information:

Both of these chemicals are mutagenic (carcinogenicity has not been substantiated, but that is no reason to treat them any less respectfully) due to their intercalating action within the DNA molecule. They are soluble, up to 10 mg./ml. in water. The recommended disposal method is incineration. The LD₅₀ for ethidium bromide (intraperitoneal injection; mice) is 20 mg./kg. of body weight. Values were unavailable for propidium iodide, but Dr. Watrud assumes they would be similar to those mentioned for the ethidium compound.

Based on this information, the description provided in your letter, and the fact that I know nothing of the equipment available or the nature of the facility in which your research will be performed, I will offer a few brief comments regarding appropriate work practices to be considered. Since these materials are available in powdered form, they are likely to be aerosolized due to electrostatic forces. Aqueous working solutions should, therefore, be prepared in advance in a controlled area. A "controlled area" may be defined as a laboratory, a portion of a laboratory, or a device such as an exhaust hood or a glove box designed for the use of highly toxic substances. I recommend that you use a negative-pressure glove box with gloves attached. The ventilation rate in the box should be at least two volume changes per hour, the internal pressure should be maintained at least at 0.5 inches of water lower than the external environment, and the exit gases should be passed through a trap or a HEPA (high-efficiency particulate air) filter. Controlled areas should be clearly marked with a conspicuous sign that says (in Spanish and English):

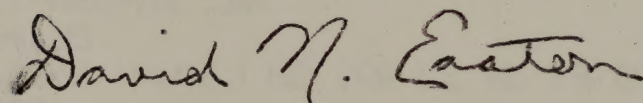
WARNING: TOXIC SUBSTANCE OR CANCER-SUSPECT AGENT IN USE: AUTHORIZED PERSONNEL ONLY.

The dry chemicals (as received from the supplier) and the aliquotted working solutions should be stored in a secured area, such as a locked freezer. Unfortunately, I have no concept of the space required to house 200,000 to 750,000 flies or the volume of 25% honey solution which has been "spiked" with the marker, but you should consider constructing a plexiglass chamber with the same negative pressure parameters and filtration requirements as those described for the glove box. This requirement is critical if there is a good possibility that the honey solution can dry out, or if excess compound is excreted by the flies. All wastes should be considered potentially contaminated and carefully containerized prior to removal and incineration.

The release of treated flies should pose no threat to the surrounding environment. Each fly's individual chemical burden is infinitesimal, and the total quantity of chemical will be effectively diluted by the actual dispersion of the population.

Thank you for the opportunity to provide some input into the planning of your project, Dr. Brenner. We appreciate and commend your health and safety concern. An excellent source of additional information is a text entitled, "Prudent Practices for Handling Hazardous Chemicals", by the National Research Council. It is available from the National Academy Press, 2101 Constitution Avenue, N.W., Washington, D.C. 20418, for about \$12.00. Feel free to contact me if you have additional questions, and please say hello to Dave Hoffman for me.

Sincerely,



David N. Easton, C.I.H.
Senior Industrial Hygienist
Safety and Health Program Management Branch

Enclosure

SS 1 /C?
USER:
FILE RTECS
PROG:
YOU ARE NOW CONNECTED TO THE RTECS FILE.
TOXICITY DATA IN NIOSH'S RTECS HAVE NOT BEEN CRITICALLY EVALUATED.

SS 1 /C?
USER:
ETHIDIUM BROMIDE
PROG:
SS (1) PSTG (1)

SS 2 /C?
USER:
PRT IL
PROG:

SI - NIOSH/SF7950000
N1 - PHENANTHRIDINIUM, 3,8-DIAMINO-5-ETHYL-6-PHENYL-, BROMIDE
RN - 1239-45-8
CC - DRUG
CC - MUTAGEN
SY - 2,7-DIAMINO-10-ETHYL-9-PHENYLPHENANTHRIDINIUM BROMIDE
SY - 2,7-DIAMINO-9-PHENYL-10-ETHYLPHENANTHRIDINIUM BROMIDE
SY - 2,7-DIAMINO-9-PHENYLPHENANTHRIDINE ETHOBROMIDE
SY - DROMILAC
SY - ETHIDIUM BROMIDE
SY - HOMIDIUM BROMIDE
MF - C21-H20-N3 .Br
MW - 394.35
WL - T B666 HKJ EJ H2 IR& LZ &E &9/26
EM - 6209

SS 2 /C?
USER:
PROPIDIUM IODIDE
PROG:
SS (2) PSTG (1)

SS 3 /C?
USER:
PRT IL
PROG:

SI - NIOSH/SF7949600
N1 - PHENANTHRIDINIUM,
3,8-DIAMINO-5-(3-(DIETHYLMETHYLAMMONIO)PROPYL)-6-PHENYL-,
DIIODIDE
RN - 25535-16-4
CC - MUTAGEN
SY - 3,8-DIAMINO-5-(3-(DIETHYLMETHYLAMMONIO)PROPYL)-6-PHENYLPHENANTHRI-
DINIUM DIIODIDE
SY - PROPIDIUM DIIODIDE
SY - PROPIDIUM IODIDE
MF - C27-H34-N4 .2I
MW - 668.45
EM - 8111



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Department of
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Southern Region
Oklahoma-Texas
Area

Agricultural Products
Quality Research
P. O. Box 267
Weslaco, Texas 78596

February 8, 1983

RECEIVED FEB 09 1983

SUBJECT: Slow release formulation of
Swormlure-4

TO: Sidney L. Vail, RL
Crop Protection Research Unit
Southern Regional Research Center
P. O. Box 19687
New Orleans, LA 70179

Thank you for your letter of January 20, 1983 concerning your unit's possible involvement in various phases of screwworm attractant (Swormlure-4) research. I am forwarding your letter and a copy of this response to Dr. O. H. Graham, Research Leader, Screwworm Research and Dr. Ralph Bram, NRP Leader for Insects Affecting Man and Animals, for consideration.

Of the four items listed in your letter, we are to some degree attempting or going to attempt to address items 1 and 3. However, your assistance may be desirable in these areas.

The development of a slow-release mechanism or formulation for Swormlure-4 is one of the greatest needs in screwworm attractants. I feel that this would aid us in determining the effectiveness and mode of action of the attractant under various conditions. Presently, we are relying on the wax formulation of the attractant in SWASS pellets, but have no workable slow release for liquid Swormlure-4 used in survey systems. Of the four possible areas, this area could be a very profitable endeavor.

Again, I will forward this to Drs. Graham and Bram for their considerable and possible suggestions for avenues of funding for this needed research. I will be talking with them and will keep you informed should anything develop.

HAROLD E. BROWN
Research Leader

cc:w/encl

R. Bram

O. H. Graham ✓

C. M. Heald

J. R. Johnston



United States
Department of
Agriculture

Agricultural
Research
Service

Southern Region
Southern Regional
Research Center

1100 Robert E. Lee Boulevard
P.O. Box 19687
New Orleans, Louisiana
70179

*Rcd's
B-4-83*

*send copy to
OH. Graham
Ralph Brown*

January 20, 1983

Subject: Slow Release Formulation for Swormlure-4

To: Harold E. Brown, Research Leader
Agricultural Products Quality Research
Post Office Box 267
Weslaco, TX 78596

Since our conversation earlier this week, I've gone over some possibilities on specific studies which could be done at Southern Regional Research Center. These suggestions may be already being worked on, but I have no way of knowing this at the present time. In cooperation with others in ARS we would hope to accomplish the following:

1. Determine the composition of volatiles in the air (odor) surrounding Swormlure-4 (or other formulations) which produce the greatest desired effect(s) upon screwworm flies.
2. Develop a system involving selective adsorption and/or other controlled release procedures that will provide for a steady release of volatile materials to optimize the effects of the lure. This would include optimization studies in the laboratory and follow-up studies as needed based on field evaluations.
3. Determine effects of temperature and moisture upon composition of the volatiles in the "odor" and apply findings to field studies.
4. Provide further support based on special skills and equipment at this Center.

We can proceed in a timely fashion if the work and reasonable funds are assigned to us. Otherwise we will be limited to providing suggestions and other limited technical support. My unit has a current funding problem which we have discussed previously. Dr. Frere and Dr. Ridgway are aware of our activities, but I do not intend to contact ARS personnel at other locations for the time being. We look for you to suggest the next step and are waiting to hear from you. Please use my memos to you as needed. This appears to be an excellent way to increase cooperation between Center scientists and ARS scientists at other locations.

Sidney L. Vaill
Sidney L. Vaill
Research Leader
Crop Protection Research Unit



United States
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Research
Service

Southern Region
Oklahoma-Texas
Area

C. G. Johnson
Agricultural Products
Quality Research
P. O. Box 267
Weslaco, Texas 78596

February 9, 1983

SUBJECT: Analysis of Supona Samples from Mexico

TO: W. H. Sudlow
Dr. Nazario Pineda-Vargas
Dr. R. E. Reichard
Dr. H. C. Hofmann
Dr. Moises Vargas-Teran

We have received Chlorfenvinphos (Supona) Standards from the U. S. Environmental Protection Agency and have determined the % supona in each sample forwarded to this laboratory from Mexico. We were unable to find a local (South Texas) laboratory that would determine supona for us.

There were two lots of material. One dated December, 1981 and the other dated January 1982. These were in 5 gram packages and we used 1 package per sample and arbitrarily assigned litters designating different samples.

Supona Samples Dated December 1981

Observation Number	Sample Number	% Supona
1	A	1.82
2	B	1.80
3	C	1.78
4	D	1.78
5	E	1.78

Summary of Data - December 1981 Samples

Number of Samples	5
Mean % Supona	1.79
Minimum Value (%)	1.78
Maximum Value (%)	1.82
Standard Deviation (%)	0.018

Supona Samples Dated January 1982

Observation Number	Sample Number	% Supona
1	A	1.41
2	B	1.28
3	C	1.78
4	D	1.74
5	E	1.75

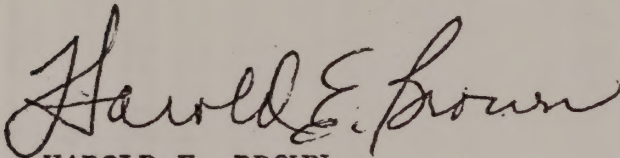
Summary of Data - January 1982 Samples

Number of Samples	5
Mean % Vapona	1.59
Minimum Value (%)	1.28
Maximum Value (%)	1.78
Standard Deviation	0.23

There was more variability in the January samples than the December samples.

I talked to Dr. O. H. Graham and he was under the opinion that 1% Supona in the powder was sufficient for treating wounds for killing screwworm larvae.

Should you have any questions please give me or Dr. Graham a call. Please distribute this to proper individuals.



HAROLD E. BROWN
Research Leader

cc:

O. H. Graham

13:06

STATISTICAL ANALYSIS SYSTEM

OBS SAMPLE PCSUPONA

A 1 1.82

B 2 1.80

C 3 1.78

D 4 1.78

E 5 1.78

SUPONA SAMPLES FROM MEXICO DATED DEC 1981

5 GRAM PACKETS LABELED 2% POWDER

(WEIGHT PERCENT)

VARIABLE N MEAN STANDARD MINIMUM VALUE MAXIMUM STD ERROR OF MEAN SLIM

PCSUPONA 5 1.7920000 0.01788854 1.7800000 1.8200000 0.0080000 8:9600000

13:08

STATISTICAL ANALYSIS SYSTEM

OBS SAMPLE PCSUPONA

A 1 1.41

B 2 1.28

C 3 1.78

D 4 1.74

E 5 1.75

SUPONA SAMPLES FROM MEXICO DATED JAN 1982

5 GRAM PACKETS LABELED 2% POWDER

FOR KILLING SCREWDRUMS

VARIABLE N MEAN STANDARD MINIMUM VALUE MAXIMUM STD ERROR OF MEAN SLIM

PCSUPONA 5 1.5920000 0.23058621 1.2800000 1.7800000 0.10312129 7.9400000



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Agricultural Products
Quality Research
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Weslaco, Texas 78596

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PCSUPONA	5	1.5920000	0.23058621	1.2800000	1.7800000	0.10312129	7.9600000

13:08 WED

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FOR KILLING SCREWORMS

(WEIGHT PERCENT)

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13:08

STATISTICAL ANALYSIS SYSTEM

VARIABLE	N	MEAN	STANDARD DEVIATION	MINIMUM VALUE	MAXIMUM VALUE	STD ERROR OF MEAN	SUM
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13:06 WED

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13:06

STATISTICAL ANALYSIS SYSTEM



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February 9, 1983

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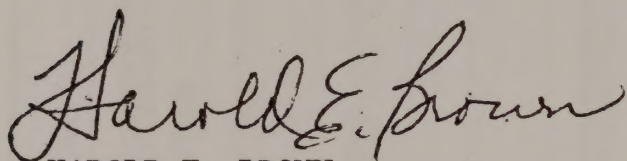
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HAROLD E. BROWN
Research Leader

cc:

O. H. Graham

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 FOR KILLING SCREWDRUMS
 (WEIGHT PERCENT)

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1	A	1.82
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PCSUPONA	5	1.7920000	0.01788854	1.7800000	1.8200000	0.0080000	8.9600000

13:08 STATISTICAL ANALYSIS SYSTEM

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 (WEIGHT PERCENT)

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VARIABLE	N	MEAN	STANDARD DEVIATION	MINIMUM	MAXIMUM	STD ERROR	SUM
PCSUPONA	5	1.5920000	0.23058621	1.2800000	1.7800000	0.10312129	7.9600000

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Weslaco, TX 78596

February 11, 1983

Dr. Frank Guillot
Research Entomologist
U. S. Livestock Laboratory
P. O. Box 232
Kerrville, Texas 78028

Dear Frank:

Please find enclosed titles and authors of the papers by Jim Mackley and Pete Peterson which were intended for the S. W. Branch Meeting.

Should you be able to work these in somehow, I sure would appreciate it.

HAROLD E. BROWN
Research Leader

Enclosures

cc:

O. H. Graham ✓

CHECK SECTION PREFERENCES:

1. Crop Protection Entomology
(including urban & industrial
entomology)
2. Ecology, Behavior, & Biometrics
(including biological control)
3. Forest & Veterinary Entomology

4. Physiology, Biochemistry,
Genetics, & Immunology
5. Systematics & Morphology
6. Forest Entomology
7. Graduate Student Award
Ballot

THIS SPACE FOR USE BY PROGRAM COMMITTEE

Date Day The Year to Received By

To
Harold

Frank:

This is the Mackley Paper.

HEP

PLEASE TYPE → NO TYPEWRITER

MAIL TO: DR. FRANK S. GUILLOT, USDA-ARS, U.S. LIVESTOCK LABORATORY, P.O. BOX 232, KERRVILLE, TX 78028.

TITLE OF PAPER (NOT TO EXCEED 15 WORDS): SWORMLURE-4: A NEW CHEMICAL

MIXTURE AS AN ATTRACTANT FOR THE SCREWORM FLY

TO BE READ BY: JAMES W. MACKLEY

(NOTE: PAPER MUST BE READ BY ONE OF THE AUTHORS.)

AUTHOR NAME(S): JAMES W MACKLEY
(FIRST) (MIDDLE INITIAL) (LAST)

HAROLD E BROWN

INSTITUTION AND ADDRESS: SCREWORM RESEARCH LABORATORY, USDA/ARS,
APARTADO POSTAL #544, TUXTLA GUTIERREZ, CHIAPAS, MEXICO
ZIP TEL NO.

Please bring your 2 x 2 slides in Kodak carousels to the meeting room.

To reserve special projection equipment, contact the program chairman no later than January 20, 1983.

CHECK SECTION PREFERENCE:

- | | |
|---|--|
| ☐ 1. Crop Protection Entomology
(including urban & industrial entomology) | ☐ 4. Physiology, Biochemistry
Genetics, & Toxicology |
| ☐ 2. Ecology, Behavior, & Bionomics
(including biological control) | ☐ 5. Systematics & Morphology |
| ☒ 3. Medical & Veterinary Entomology | ☐ 6. Forest Entomology |
| | ☐ 7. Graduate Student Award
Contest |

THIS SPACE FOR USE BY PROGRAM COMMITTEE.

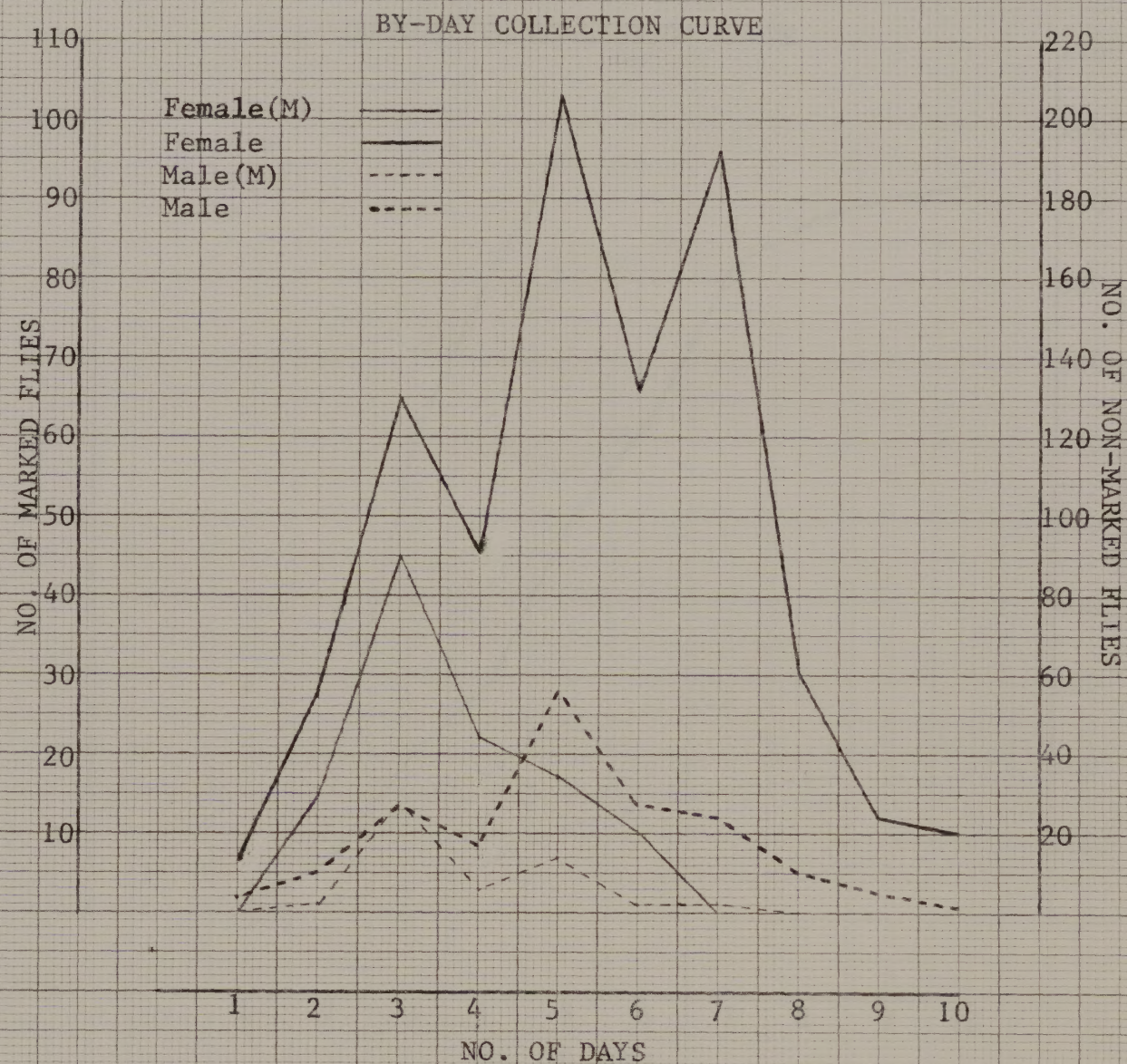
Section	Date	Day	Time	Paper No.	Received	Misc.
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Frank:
This is Peter's copy.
HCB.

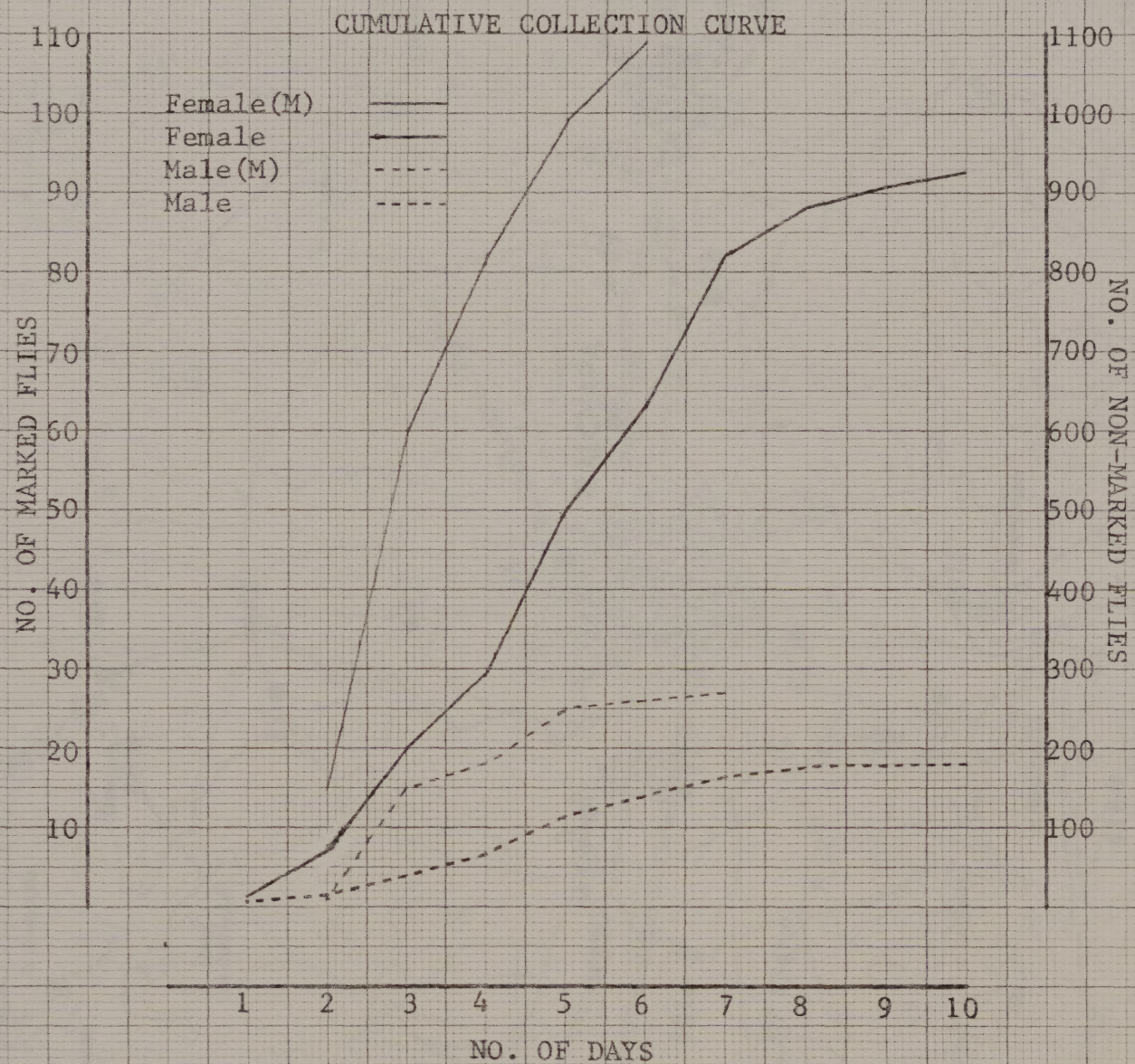
Longevity and Mating
Capacity of Male Screwworm
(Diptera: Calliphoridae)

Richard D. Peterson,
Agustin Ocampo-Candido, and
H. DelVar Petersen

Screwworm Research, USDA/ARS
Apartado Postal #544,
Tuxtla Gutierrez, Chiapas
Mexico, and USDA,ARS
Oklahoma-Texas Area, Southern Region
College Station, Texas 77840



1 10-Day Trial
 February 12-21, 1983
 Marked Fly ground-release February 11
 Regular Releases February 8 and 14
 (M) = Marked Flies



1 10-Day Trial
 February 12-21, 1983
 Marked Fly ground-release - February 11
 Regular releases - February 8 and 14
 (M) = Marked Flies

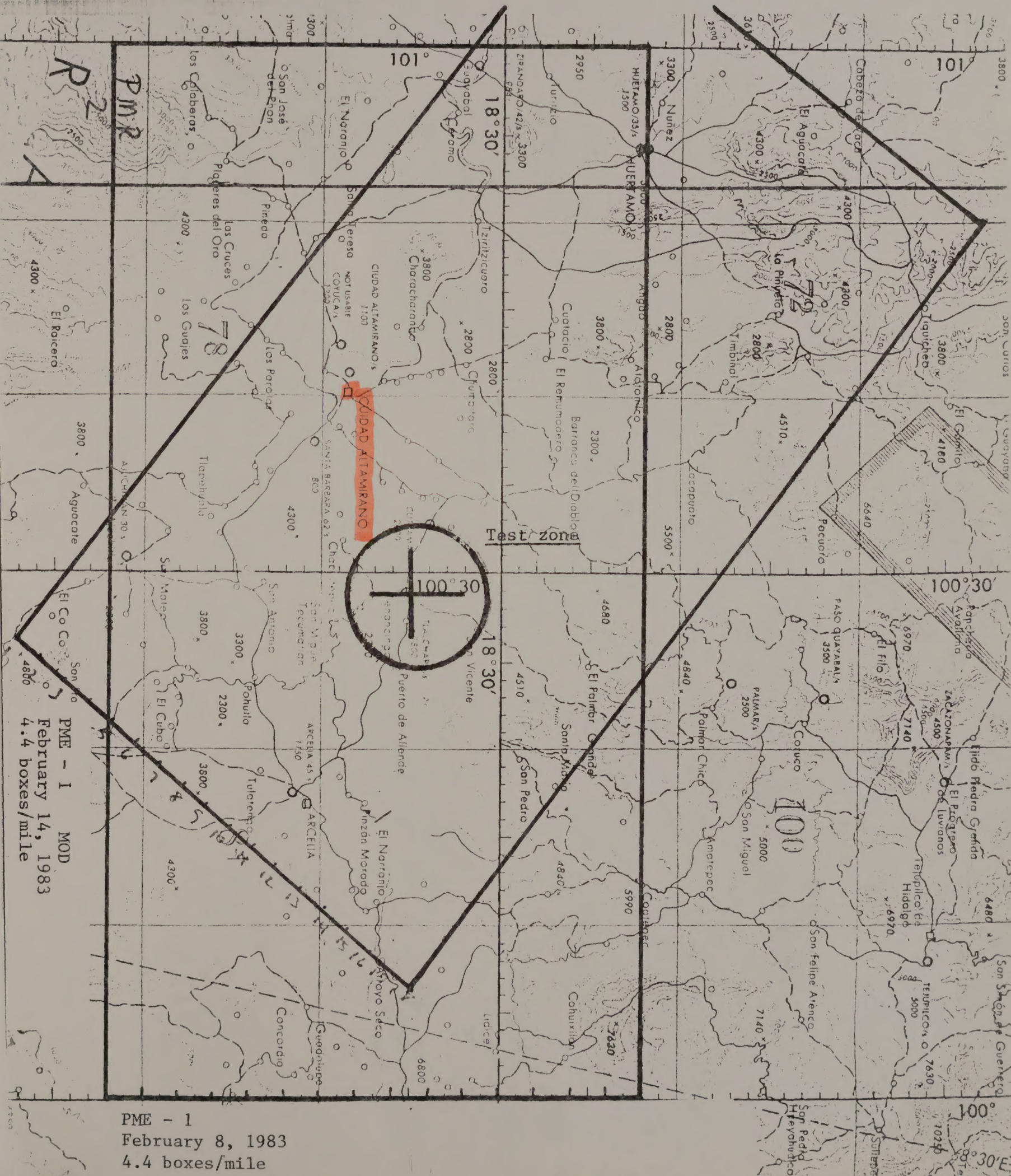


DAILY SCREWORM COLLECTION

DAY	1		2		3		4		5		6		7		8		9		10	
TRAP	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F
# 1	0	0	0	0	0	2	0	1(1	0	1	0	0	0	0	-	-	0	1	0	1
# 2	0	0	0	2	3(1	11(4	2	10(3	12	29(2	3	17(2	2(1	28	1	6	0	1	0	6
# 3	0	0	0	0	1(1	0	0	0	0	1	0	4	0	8	1	3	0	0	0	1
# 4	*1	**6	3	14	1	14(3	0	5(2	3	*8(1	3	26(2	9	41	1	14	0	0	0	0
# 5	0	1	0	3	0	1(1	0	0	0	1	1	1	0	10	3	10	0	0	0	0
# 6	0	0	2	9	2	1	0	1	*4	*11(1	1	11	2	20	0	0	0	4	0	*3
# 7	0	0	0	0	0	3(2	0	0	*1	4	0	0	0	0	0	0	0	0	0	0
# 8	0	2	0	1	0	4	-	-	0	1	0	2	0	3	0	1	1	0	0	1
# 9	0	0	0	1	0	5(1	2(1	3	*3(1	11(2	0	1	0	2	0	0	0	0	0	0
# 10	0	0	1	3(1	3(1	12(4	1	1	0	0	5	5(1	8	24	1	5	3	3	0	1
# 11	0	0	0	0	0	5(1	0	0	0	4	0	2	0	0	0	0	0	0	0	0
# 12	0	0	0	1	0	0	0	3	1	9(1	0	0	0	6	0	2	0	1	0	0
# 13	0	0	0	7(6	2(1	4(3	1	8(1	0	6(1	1	12(2	0	0	0	0	0	0	0	0
# 14	0	0	0	3	9(6	34(13	6(2	18(4	4	14(4	0	2	0	0	0	0	0	1	0	0
# 15	0	0	0	1	0	1	0	1(1	0	3(1	0	1	0	1	0	2	0	1	1	0
# 16	0	2	0	1	0	0	0	0	0	0	0	0	0	0	0	*1	0	0	0	1
# 17	1	0	0	4	0	*3	0	0	0	1	0	1	0	0	0	0	0	0	0	0
# 18	0	0	0	0	0	0	0	2	0	1	0	0	0	5	0	0	0	0	0	2
# 19	0	1	5(1	14(8	10(3*	46(10	4	*33(10	11(4	53(3	3(1	8(2	0	2	0	2	0	0	0	2
# 20	0	0	0	1	5(1	8(2	1	4	2	3	0	3	-	-	0	0	0	0	0	0
# 21	1	0	0	2	0	3(1	0	3	1(1	6	0	4(1	0	1	0	1	0	0	0	1
# 22	0	0	0	0	3	6	1	6	2	6	9	36	3	38	2	12	0	2	0	0
# 23	0	1	0	2	1	4	1	3	9	22	0	0	1	0	1	0	0	2	0	1
# 24	0	0	0	2	1	8	1	11	10(1	28(1	2	5	0	3	0	2	1	8	0	0
	3	13	11	71	41	175	20	113	63	223	28	141	25	192	10	61	5	24	1	20
			(1)	(15)	(14)	(45)	(3)	(22)	(7)	(17)	(1)	(10)	(1)							
	*	**				**		*	***	**						*				*

NOTE: Each asterisk*- represents one wild fly
 The numbers in parenthesis-(#)- represent marked flies
 Trapping took place February 12-21, 1983

2 grids were employed to treat the area where the test took place
the date of the dispersal and the amount of boxes dropped are indicated on map



Dr. Graham



United States
Department of
Agriculture

Agricultural
Research
Service

Southern Region
Oklahoma-Texas Area

Screwworm Research
P. O. Box 986
Mission, TX 78572

February 15, 1983

Dr. L. Levenbook
National Institutes of Health
Building 6, Room 137
Bethesda, Maryland 20205

Dear Dr. Levenbook:

Thank you for your interesting comments on the growth and behavior of Calliphora larvae. While I must admit that I had hoped that you would supply some easy answers to a complex problem, I was not suprised to read that you have found that growth may be influenced by several factors.

Unfortunately we do not have very much factual evidence to present but I believe that most of the people who have reared screwworms over a period of years would agree that the larvae in a "critical mass" (we refer to them as "pockets") grow faster than larvae which stray from the mass. The changes which occur in the color and consistency of the medium support your supposition that digestive enzymes are excreted by the feeding larvae and then ingested along with food material from the medium. In a mass-rearing plant larvae do leave an overcrowded vat prematurely and the operators have to achieve a very delicate balance between too few, which would be inefficient, and too many, which probably contributes to an expanded crawl-off period and a wider range of larval sizes.

Sincerely,

O. H. Graham

O. H. GRAHAM
Location/Project Leader

cc: (with incoming)

R. A. Bram
H. E. Brown
R. L. Harris
R. D. Peterson
H. C. Hofmann
Ron Lowe



DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

National Institutes of Health
Bethesda, Maryland 20205

Mailing Address:

National Institutes of Health
Building 6, Room 137
Bethesda, Maryland 20205

January 21, 1983

Dr. O. H. Graham
USDA, ARS
Screwworm Research Project
P.O. Box 986
Mission, Texas 78572

Dear Dr. Graham:

I apologize for my belated reply to your letter of January 5th. I fully understand your problem since, on a much smaller scale of breeding, we encounter a similar lack of larval synchrony despite the fact that eggs are collected over an hour's laying period. Personally, I doubt whether this has anything to do with ecdysterone or hormone secretion as the larvae have plenty of food available even though they don't seem to utilize it at the same rate. About the only clue I have is that this lack of synchrony may have to do with the feeding and rate of growth of first instar larvae. We have observed that when our Calliphora larvae are raised under aseptic conditions on a semi-defined medium, several newly hatched individuals wander about for many hours during which time they appear to feed and grow very little if at all. At the same time other larvae appear to aggregate and collectively attack the diet at some particular location. These latter larvae are clearly larger, and ecdyse to the 2nd stage well ahead of the wandering ones. It would seem as though a "critical mass" of contiguous larvae are more efficient in digesting food than a solitary larva alone. This in turn may have to do with the fact that blowfly -- and I suspect, without knowing -- also screwworm larvae, feed by secreting digestive enzymes externally, and sucking up the digested products of proteolysis.

One further point, with which I am sure you are familiar, is that overcrowding the larvae results in a wider range of fully grown larval sizes, and a corresponding variation in adult fly weights, than when there is a lower larval density. Could this possibly be a factor in your rearing?

I regret that my comments are not more helpful, but the problem you raise is not one I am really competent to answer.

Yours very sincerely,

L. Levenbook
Laboratory of Physical Biology
National Institute of Arthritis,
Diabetes, and Digestive and Kidney
Diseases

LL:bjm



United States
Department of
Agriculture

Agricultural
Research
Service

Southern Region
Oklahoma-Texas Area

Screwworm Research
P. O. Box 986
Mission, TX 78572

January 5, 1983

Dr. L. Levenbook
Laboratory of Physical Biology
National Institutes of Health
Bethesda, MD 20205

Dear Dr. Levenbook:

I have read with great interest your paper with Dr. Shaaya on the feeding and pupariation of C. vicina larvae (J. Insect Physiol. 28: 683-688) and have speculated on the possible application of your findings to our research on the mass rearing of screwworm larvae. USDA, in cooperation with Mexican government agencies, operates a rearing plant at Tuxtla Gutierrez, Chiapas which routinely produces 300 to 500 million screwworm pupae per week. When newly colonized strains are first placed into mass production we often see a great deal of variation in the time that larvae spend in the rearing medium and the size of the crawl-off larvae. Many larvae leave the medium too soon and produce small flies of reduced vigor. Others do not crawl off of the rearing vats at the end of the normal rearing cycle (about 6 days) and are lost in the discarded medium.

Obviously program efficiency would benefit tremendously if larval nutrition and physiology could be deliberately manipulated so that all larvae attain a size of 60-75 mg at close to the same time (plus or minus 24 hr). Do you see any possibility that one or more of the subtle nutritional or hormonal factors which influence larval growth and puparium formation could be regulated so as to approach this ideal? We would greatly appreciate any comments you might care to make.

Sincerely,

O. H. GRAHAM
Location/Project Leader

cc:

R. A. Bram
H. E. Brown
R. L. Harris
R. D. Peterson
H. C. Hofmann
R. Lowe



United States
Department of
Agriculture

Agricultural
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Service

Southern Region
Oklahoma-Texas Area

Screwworm Research
P. O. Box 986
Mission, TX 78572

February 15, 1983

Dr. R. A. Hoffman
Associate Area Director
USDA, ARS, SR, Oklahoma-Texas
Area
P. O. Drawer EC
College Station, Texas 77841

Dear Bob:

Time flies - it doesn't seem all that long since you passed through Kerrville en route to Stoneville from Corvallis. The 26 plus years that I have known you have been good ones for me and I'm pretty sure you feel the same way about the time we've spent in ARS. We have had the privilege of being assigned to the most important program in ARS and working with its finest people. We didn't solve all of the problems assigned to us, but we can take pride in the progress that has been made.

I can't help but feel that you're too young to retire but then you've worked a lot harder than I have and really have earned a chance to relax a little, rest a little, and to just plain enjoy life. And that's exactly what all of us are wishing for you - the finest years of your life. All the best for you and Tobe.

Sincerely,

O. H. GRAHAM
Location/Project Leader

BOB HOFFMAN'S RETIREMENT

The following is enclosed for Dr. Hoffman's retirement:

Letter to Bob Hoffman (UNFOLDED)	<u>✓</u>
Contribution for gift (\$ <u>20⁰⁰</u>)	<u>✓</u>
*Cost of dinner - \$12.00 per person (Includes Tax & Gratuity)	<u>No</u>
Total	<u> </u>

Make checks payable to Ella Flynt and return
to the following address by February 24, 1983.

Ella Flynt
Oklahoma-Texas Area Office
P. O. Box EC
College Station, TX 77841

*A cash bar will be available from 6-7pm.

THE FOLLOWING IS A SUMMARY

The following is a summary of the information received from the following sources:

1	Letter to the Hon. Mr. [Name] (1950)
2	Constitution for the [Name] (1950)
3	*Letter to the Hon. Mr. [Name] (1950)
4	(Letter to the Hon. Mr. [Name])
5	Total

Note: The above figures are for the year 1950 and are subject to change as more information is received.

Very truly yours,
[Signature]
[Name]
[Address]
[City, State, Zip]

at such time as will be available from the [Name]



United States
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Agriculture

Agricultural
Research
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National
Program
Staff

Shaham
Beltsville, Maryland
20705

RECEIVED MAR 03 1983

February 16, 1983

SUBJECT: Trip Report - Screwworm Research Unit, Metabolism and Radiation
Research Laboratory

TO: D. B. Laster, ADA, AHNPS

Location: Fargo, North Dakota

Dates: February 7-10, 1983

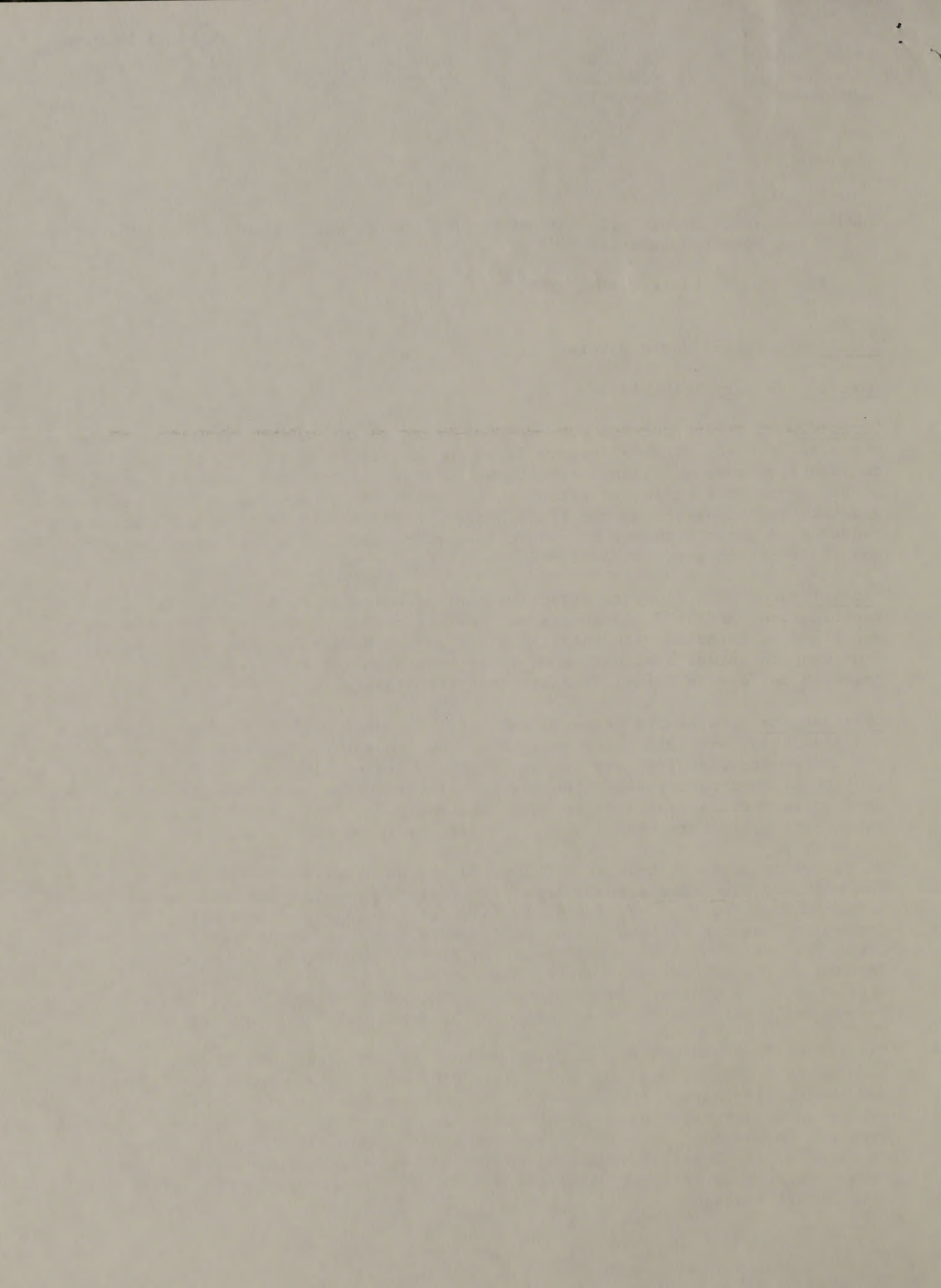
Purpose: To review current screwworm research in the newly created Screwworm Research Unit and other screwworm research activities throughout the laboratory; to identify those additional resources necessary to increase research activity on screwworm attraction; to establish the direction of new research on screwworm genetics made possible by the FY 83 budget increase; and to coordinate screwworm research at Fargo with the Screwworm Research Unit, Tuxtla Gutierrez, Mexico, and the research needs of APHIS-VS.

Participants: NPL, Insects Affecting Man and Animals, F. E. Smith, Chief Staff Veterinarian, APHIS-VS, International Operations and Screwworm Staff; the visit was timed to coincide with the site visit of the National Academy of Sciences, Committee on Animal Health Team on long-range planning for diagnosis and research on foreign animal diseases and ectoparasites.

Observations: Screwworm research facilities at MRRL are of the highest quality and consistent with all other aspects of the laboratory. It was obvious that the long-range planning team was extremely impressed with the facilities, the quality of ongoing research, the positive interactions among scientists, and the array of expertise available at this location. Dr. Smith and I also found the entire scientific atmosphere at the laboratory to be exhilarating.

Screwworm research at MRRL is not limited to the Screwworm Research Unit, but on the contrary, includes components of the Insect Physiology and Biochemistry Research Unit and the Insect Genetics and Cell Biology Research Unit. The Screwworm Research Unit maintains at least 20 lines of screwworms for use of all investigators. Personal research of the Project Leader, C. J. Whitten, is focused on the discovery of genetic sexing mechanisms (all male strain), flower attractants (in cooperation with R. A. Flath, WRRC and the Tuxtla Gutierrez Screwworm Research Unit), and electrophoretic analysis of isofemale lines.

The Insect Physiology Research Unit concentrates on screwworm attraction where L. Hammack (100% of time) and G. Pomonis (20% of time) are working to identify and eventually characterize aromatic factors that attract screwworms; attractants currently under investigation include dried blood components, wound bacteria emanations, and limited work on female mating stimulants. Other research by the Physiology Unit which has potential for application to the screwworm program includes endocrine control of reproduction and behavior, and nutritional research.



The Genetics Research Unit emphasizes research on screwworm genetics where L. E. LaChance (30% of time), NTA for Insect Genetics, addresses the genetic characterization of isofemale lines, mating preference tests with isofemale lines, genetic mapping when the opportunity arises, and overall guidance and advice on screwworm genetic research. Other activities of the Genetics Research Unit having relevance to the screwworm include isozyme and monoclonal antibody studies, molecular analyses, embryo mitotic mechanisms and germplasm preservation. This unit also supports the research on bacterial attractants through cooperative agreement with M. Bromell, North Dakota State University.

Recommendations:

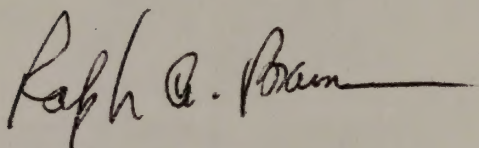
1. The need for additional research on screwworm attraction is obvious to all associated with the screwworm program. However, it does not now appear that research on the chemistry of attraction per se is slowing progress, but instead that the bioassay of the various chemical components is the bottleneck. Much of Dr. Hammack's time is devoted to the necessary, but routine, chores of preparing screwworms and olfactometers for analysis of different attractant components, detracting from creative input to attraction and behavior research.
 - o It is, therefore, recommended that a PFT technician be hired to assist in the area of screwworm attraction, permitting greater scientific productivity in this area by both Dr. Hammack and Dr. Pomonis.
 - o An effort should also be made to acquire sufficient quantities of attractant components to permit field evaluation at Tuxtla Gutierrez simultaneously with olfactometer tests at Fargo. Coordination will be needed with O. H. Graham and H. E. Brown with appropriate followup.

Dr. M. Bromell's work on bacterial attractants has been most productive and shows promise of providing a key to wound attraction. Unfortunately, her status with the University may be in jeopardy without additional cooperative agreement support.

 - o It is, therefore, recommended that consideration be given by the Screwworm Research Unit to provide the additional support necessary to maintain the active pursuit of this line of research.
2. The FY 83 budget increase for screwworm genetics provides the opportunity to expand fundamental studies at Fargo which will be of greatest value to the screwworm program. Since Dr. LaChance is now devoting 1/3 of his time to screwworm genetics, since a post doctoral student will shortly be hired to study polytene chromosomes, and since the Screwworm Research Unit at Tuxtla Gutierrez has just hired a geneticist to replace Dr. McInnis, the time seems propitious to fully investigate the question of heterosis as suggested by the recent Lincoln-Eden report.
 - o It is, therefore, recommended that a new SY be established in the Screwworm Research Unit to study hybrid vigor with eventual coordination with research and methods development at Tuxtla Gutierrez.

- o First priority for this position should be offered to a scientist at the Screwworm Research Unit, Tuxtla Gutierrez, who is ready to return to the United States.

3. The addition of Dr. Whitten to the MRRL staff has fostered closer coordination and cooperation between the fundamental screwworm research at Fargo and the more field oriented activities at Tuxtla Gutierrez. He should be commended for this progressive accomplishment. The cooperation of the Genetics Research Unit and Physiology Research Unit add strength to the total research effort and Drs. Gassner and Nelson deserve acknowledgement and support for their long-term commitment.



RALPH A. BRAM
National Program Leader
Insects Affecting Man & Animals

cc:

T. J. Army
P. J. Fitzgerald
K. L. Lebsock
C. H. Schmidt
C. J. Whitten
G. Gassner
D. R. Nelson
L. E. LaChance
✓ O. H. Graham
H. E. Brown
F. E. Smith, APHIS

22nd February 1983

Dr. Hugh Graham
US Department of Agriculture
ARS
PO Box 986
Moore Air Base
Mission
Texas 78572

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Middlesex House
34-42 Cleveland Street
London W1P 5FB
Telephone: 01-580 9327
Telex: 261661

Dear Dr. Graham,

I am writing on behalf of Dr. Dennis Jacobs under whose authorship we are producing "Equine Parasitology". This book will be one volume in a series on parasitology.

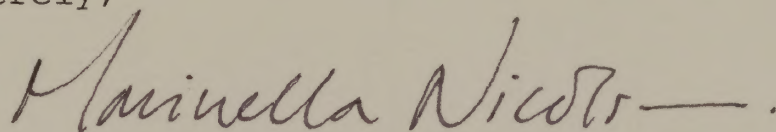
"Equine Parasitology" will be highly illustrated with colour photographs of the parasites and the lesions they cause. A short text will outline the diagnostic features of the disease together with some information on the life-cycle of the parasite. The bias will be towards the clinician as opposed to the zoologist, but we intend the book to reach both students and practitioners.

I am now collecting the photographs, and have been quite overwhelmed by the response from our contributors all over the world. We have in fact, collected most of the photographs but there are some which still elude me. I was hoping you might be able to help me with colour transparencies of the screwworm, adult and larva, together with pictures of the lesions it causes. I am most interested in pictures showing the lesion in horses but if you only have pictures of cattle I would accept them.

In return for your permission to publish the photograph, we give an acknowledgement in its caption and pay your expenses for film, post etc. I would prefer to use the original and will supply you with copies until the original is returned after publication.

I do hope you would like to contribute to our book. I look forward to hearing from you and send my thanks in advance.

Yours sincerely,



Marinella Nicolson (editor)

O. N. Graham



COMISION MEXICO-AMERICANA PARA LA ERRADICACION
DEL GUSANO BARRENADOR DEL GANADO

FEBRERO 22, 1983

MR. HAROLD E. BROWN
RESEARCH LEADER

I APPRECIATE YOUR INFORMATION REGARDING THE " SUPONIA
ANALYSIS"

I WILL DISCUSS THIS INFORMATION WITH THE METHODS
DEVELOPMENT PERSONNEL.

THANKS.

[Signature]
DR. NAZARIO PINEDA VARGAS
DIRECTOR GENERAL

*MPV

3/7/83

*Hugh:
I received this response
from Dr. Pineda Today
HEB*



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Southern Region
Oklahoma-Texas Area

Screwworm Research
P. O. Box 986
Mission, TX 78572

February 24, 1983

SUBJECT: Application for a Sabbatical Position -
Marvin D. Coffey

TO: Dr. R. A. Bram
USDA, ARS, NPS, LVS
Bldg. 005 Room 211
BARC-WEST
Beltsville, MD 20705

Reference is made to the exchange of correspondence last November between you and Dr. Coffey. In principle, I favor the establishment of a sabbatical position in the Screwworm Research Laboratory because I believe that a well-trained entomologist who was dedicated to research could complete one or more limited items of screwworm research in a 12-months' period (but not in a shorter period - 12 months is probably the absolute minimum). However, before I could recommend the appointment of a particular individual, I would want to assume myself that he/she (1) was seriously interested in completing publishable research on our species, (2) would be able to adapt to our special set of living and working conditions, and (3) would not require such a large amount of assistance from other professionals at Tuxtla as to be a burden rather than a help. We would almost certainly need to interview an applicant before we could commit ourselves.

It appears to me that an appointee would have to obtain a collaborator's appointment and an official passport before travel to Mexico could begin. With a collaborator's appointment it would then be possible for ARS to pay the appointee's travel costs and a per diem allowance that would cover part, perhaps almost all, of the appointee's living costs in Tuxtla Gutierrez.

Dr. O. H. GRAHAM
Location/Project Leader

cc:

J. R. Johnston
O. R. Garcia



United States
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Southern Region
Oklahoma-Texas Area

Screwworm Research
P. O. Box 986
Mission, TX 78572

February 28, 1983

SUBJECT: Analyses of Supona Samples from Mexico

TO: Dr. Harold E. Brown

Thank you for sending me a copy of the information included in your memorandum of February 9 on this subject. I know that this and similar information you have supplied in the past is also greatly appreciated by Commission and APHIS program managers. Lotsa bucks are involved and this type of analysis is not available from any other source.

Hugh

O. H. GRAHAM
Location/Project Leader

cc:

J. R. Johnston (w/copy of incoming)

R. A. Bram (w/copy of incoming)

O.G. Johnson



United States
Department of
Agriculture

Agricultural
Research
Service

Southern Region
Oklahoma-Texas
Area

Agricultural Products
Quality Research
P. O. Box 267
Weslaco, Texas 78596

February 9, 1983

SUBJECT: Analysis of Supona Samples from Mexico

TO: W. H. Sudlow
Dr. Nazario Pineda-Vargas
Dr. R. E. Reichard
Dr. H. C. Hofmann
Dr. Moises Vargas-Teran

We have received Chlorfenvinphos (Supona) Standards from the U. S. Environmental Protection Agency and have determined the % supona in each sample forwarded to this laboratory from Mexico. We were unable to find a local (South Texas) laboratory that would determine supona for us.

There were two lots of material. One dated December, 1981 and the other dated January 1982. These were in 5 gram packages and we used 1 package per sample and arbitrarily assigned litters designating different samples.

Supona Samples Dated December 1981

Observation Number	Sample Number	% Supona
1	A	1.82
2	B	1.80
3	C	1.78
4	D	1.78
5	E	1.78

Summary of Data - December 1981 Samples

Number of Samples	5
Mean % Supona	1.79
Minimum Value (%)	1.78
Maximum Value (%)	1.82
Standard Deviation (%)	0.018

Supona Samples Dated January 1982

Observation Number	Sample Number	% Supona
1	A	1.41
2	B	1.28
3	C	1.78
4	D	1.74
5	E	1.75

October 1, 1954

Summary of Progress, Southern Region, October 1954

1. J. H. Campbell
2. R. L. Metcalf
3. W. H. Anderson
4. J. E. Davis
5. J. H. Campbell

The following summarizes the progress of the Southern Region during the month of October, 1954. The work was divided into five main areas: (1) general entomology, (2) plant pathology, (3) insect physiology, (4) insect behavior, and (5) insect control. In general entomology, the work was divided into two main areas: (1) general entomology, and (2) plant pathology. In plant pathology, the work was divided into two main areas: (1) general entomology, and (2) insect physiology. In insect physiology, the work was divided into two main areas: (1) general entomology, and (2) insect behavior. In insect behavior, the work was divided into two main areas: (1) general entomology, and (2) insect control.

General Entomology, October 1954

Number	Species	Count
1	A	1.00
2	B	1.00
3	C	1.00
4	D	1.00
5	E	1.00

Plant Pathology, October 1954

Number	Species	Count
1	A	1.00
2	B	1.00
3	C	1.00
4	D	1.00
5	E	1.00

Insect Physiology, October 1954

Number	Species	Count
1	A	1.00
2	B	1.00
3	C	1.00
4	D	1.00
5	E	1.00

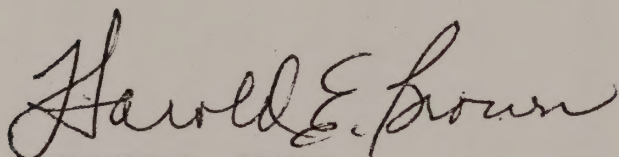
Summary of Data - January 1982 Samples

Number of Samples	5
Mean % Vapona	1.59
Minimum Value (%)	1.28
Maximum Value (%)	1.78
Standard Deviation	0.23

There was more variability in the January samples than the December samples.

I talked to Dr. O. H. Graham and he was under the opinion that 1% Supona in the powder was sufficient for treating wounds for killing screwworm larvae.

Should you have any questions please give me or Dr. Graham a call. Please distribute this to proper individuals.



HAROLD E. BROWN
Research Leader

cc:

O. H. Graham

STATISTICAL ANALYSIS SYSTEM

13:06 WEDNESDAY, FEBRUARY 2, 1983

OBS SAMPLE PCSUPONA

1 A 1.82
2 B 1.80
3 C 1.78
4 D 1.78
5 E 1.78

SUPONA SAMPLES FROM MEXICO DATED DEC 1981
5 GRAM PACKETS LABELED 2% POWDER
FOR KILLING SCREWORMS
(WEIGHT PERCENT)

13:06 WEDNESDAY, FEBRUARY 2, 1983

VARIABLE	N	MEAN	STANDARD DEVIATION	MINIMUM VALUE	MAXIMUM VALUE	STD ERROR OF MEAN	SUM	VARIANCE	C.V.
PCSUPONA	5	1.79200000	0.01788854	1.78000000	1.82000000	0.00800000	8.96000000	0.00032000	0.998

STATISTICAL ANALYSIS SYSTEM

13:08 WEDNESDAY, FEBRUARY 2, 1983

OBS SAMPLE PCSUPONA

1 A 1.41
2 B 1.28
3 C 1.78
4 D 1.74
5 E 1.75

SUPONA SAMPLES FROM MEXICO DATED JAN 1982
5 GRAM PACKETS LABELED 2% POWDER
FOR KILLING SCREWORMS
(WEIGHT PERCENT)

13:08 WEDNESDAY, FEBRUARY 2, 1983

VARIABLE	N	MEAN	STANDARD DEVIATION	MINIMUM VALUE	MAXIMUM VALUE	STD ERROR OF MEAN	SUM	VARIANCE	C.V.
PCSUPONA	5	1.59200000	0.23058621	1.28000000	1.78000000	0.10312129	7.96000000	0.05317000	14.484

✓ O. H. Graham

02/28/83

Proposal

by H. E. Brown

Title: Determination of screwworm attractant fractions
from umbilical chord and navel tissue of new born animals

Background:

It has been recognized by ranchers, old screwworm researchers, veterinarians and control program persons, as well present day scientists that the navel tissue of new born animals (calves, lambs, kids, etc) serves as a prime oviposition and investment site for screwworms. This may be due to natural components of the tissue or products due to natural (autolytic) or bacterial decomposition. Isolation of fractions from this tissue, that provide a certain degree of attractiveness and the identification of these components may provide information concerning the enhancement of swormlure-4 and new attractants for use in SWASS, survey systems and as oviposition stimulants.

Plan of Work:

Umbilical chords and navel tissue will be collected from new born calves at a local dairy. This source has a herd of 750 adult cows which should provide an adequate supply of tissue.

Arrangements have been made with the management of the dairy to allow their personnel to collect the tissue when born, place the tissue in containers provided by ARS and give us a call. Those samples collected at night, are to be called in shortly after 8:00A.M. daily, and held in a refrigerator until we pick them up. This dairy has agreed to provide new born bull calves for navel tissue collections when, and if, needed.

Tissue will be transported to the Agricultural Products Quality Research Weslaco and various extracts and preparations will be prepared. Each of the steps of work will depend on results from previous efforts.

Initially, preparations of tissue will be made by extracting, individually and sequentially with various solvents with increasing polarity. (i.e. hexane, methylene chloride, acetone, methanol) and with solvent mixtures and pairs. Other preparations using freeze dried materials and acetone powders will be used as test fractions.

Samples, once prepared, will be properly stored until they can be transported to Tuxtla Gutierrez, Chiapas, Mexico for bioassay. Bioassay of fractions for attractancy and oviposition stimulation will be conducted with fertile females in the laboratory using trap jar techniques and with gravid females for oviposition attractants. Techniques of these assays will be worked out during the initial stages.

Those fractions that yield positive results will be subjected to field evaluations, first at the mass-rearing plant in Tuxtla Gutierrez, Chiapas then in the field in areas having wild and released (sterile) fly populations. These fractions will be subjected to further chemical/physical fractionation and

Transmitted electronically to Ralph Bram
02/28/83

sub-fractions will be bioassayed in a similar manner. This process will be repeated until classes of compounds, or individual materials, have been identified. Results from these studies may provide necessary information for formulating a new attractant.

Facilities:

Facilities designated for these studies will be: (1) the analytical and preparative laboratories of the Agricultural Products Quality Research Unit in Weslaco, Texas; (2) the Screwworm Research Unit's fly secure laboratory in Tuxtla Gutierrez Chiapas; (3) The Methods Development Section at the Mexico-American Commission's mass-rearing facility at Tuxtla Gutierrez Chiapas (if needed); and (4) the area around the facility for bioassay purposes.

Personnel:

Harold E. Brown, ARS, Weslaco, TX.
James E. Mackley, ARS, Tuxtla Gutierrez, Chiapas, Mexico
William H. Sudlow, APHIS, VS, Mission, TX.
Paul E. Thompson, ARS, Weslaco, TX.
Melquiades Leal, ARS, Mission, TX.
Jose Garcia, ARS, Mission, TX.
Maria G. Rodriquez, Mexican-American Commission, Mission, TX
Arturo Vela, Mexican-American Commission, Mission, TX

Harold E. Brown will serve as project coordinator with the assistance of W. H. Sudlow and James E. Mackley. This group, in concert, will plan sequential steps in the approach. This approach will be coordinated with O. H. Graham and others at Tuxtla Gutierrez by James E. Mackley.

W. H. Sudlow will be assigned blocks of time ranging up to 10 days in duration, initially, to travel to Tuxtla Gutierrez for performing bioassays and making observations on various fractions, samples and/or materials as needed in conjunction with James E. Mackley. Jose Garcia and Melquiades Leal will be used for collecting, preparing and handling samples and performing bioassays as needed, when available. Paul E. Thompson, Maria G. Rodriquez will be responsible for chemical/physical fractionation and sample preparation. Arturo Vela will be responsible for collecting samples and assisting Thompson and Rodriquez in sample preparation.

Schedule:

Work has been initiated and will be accelerated as soon as all parties are contacted and are in agreement with this proposal. W. H. Sudlow's time will be allocated, when available and mutually agreeable, in blocks of days in order to accomplish specific bioassays. Thompson, Rodriquez and Vela will spend full time in sample preparation and fractionation.

James E. Mackley, O. H. Graham and others at Tuxtla Gutierrez will be consulted and proper arrangements made with time allowed for adequate preparation of bioassay material to be used in experiments.

INSTRUCTIONS FOR PREPARING ABSTRACT

1. Abstracts will be photographed just as you submit them, so please follow the suggested format.
2. Abstracts should be typed in a 3 x 7 inch space, such as provided below. Letters should not touch the lines. Single space all typing, leaving no top or left margins.
3. Use an electric typewriter with a good ribbon; make neat corrections. Elite type is preferred. Use black ink for symbols not on your typewriter.
4. Your Abstract should be organized as follows:
 - a. Title- use CAP'S; check to see that Transmittal Form and Abstract titles match; when using a Latin name, include the common name also; see example below.
 - b. Authors- include department and institutional affiliation.
 - c. Specific objectives- include brief statement unless clear from the title.
 - d. Methods- include a brief description, if pertinent.
 - e. Results- provide a clear, specific summary.
 - f. Conclusions-
 - g. Acknowledgements- if desired.
5. Send the ORIGINAL PLUS ONE COPY of Abstract along with the Transmittal Form to the Program Officer.

Example of title lines:

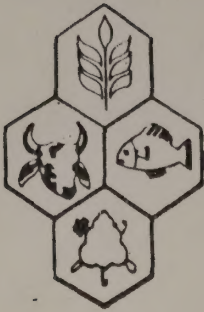
ECOLOGICAL AND BEHAVIORAL DETERMINANTS OF APPARENT MONOGAMY IN EASTERN BLUEBIRDS
(Sialia sialis)

Patricia Adair Gowaty, Dept of Zoology, Clemson University

BEHAVIORAL RESPONSES OF NATURAL POPULATIONS OF SCREWORMS, Cochliomyia hominivorax (COQ.), TO STERILE FLY RELEASE

Robert L. Mangan, USDA, ARS, Tuxtla Gutierrez, Chiapas, Mexico

Rates of egg deposition have been recorded for target populations of screwworms in eradication programs and field experiments in Florida, various Caribbean islands, and subtropical and tropical regions of Texas and Mexico from 1952 to the present. Most population monitoring has been performed for evaluation of candidate strains of flies for mass production. Rates of egg mass sterility are used as performance criteria. Comparison of pre- and post-release oviposition data, or data from treated and nontreated adjacent areas, indicates that native populations may experience significant interference by sterile flies. This apparently results in temporary reduction in the rate of oviposition in the target population. This effect has been noted in tests when target populations have never been treated with sterile flies and when release density was many times greater than the estimated natural population density.



MINISTERIE VAN LANDBOUW, VEETEELT, VISSERIJ & BOSBOUW

P.O.B. 1016

Tel: 82440

AFDELING VETERINAIRE INSPECTIE

Onderwerp: "Screw worm".

Ons kenmerk 45/026

Biilage(n)

Paramaribo 28 februari 1983

To Mr. Sam Rawlins
Department of Microbiology
U.W.I.
Mona
Kingston 7,
Jamaica

Dear Sam,

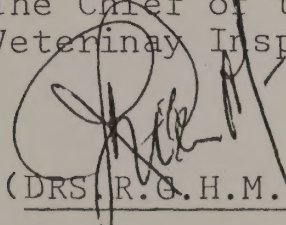
I trust you and your family are well.
On a rather new beefcattle farm (Tibiti) in the south of Surinam we have a serious screw worm problem, which we would like to control.

As such we would like to try "Swass" or bait stations. The area is $\pm$ 500 ha and isolated, far from other farms, so baits would be put around the area.

Therefore we would like some more information about the medium, where we can order it, the costs etc.

I hope to hear from you very soon.

The Chief of the
Veterinary Inspection,


(DRS. R.G.H.M. LIEUW A JOE)

cc: Drs. Moll

